10/697266

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=> d his

     (FILE 'HOME' ENTERED AT 14:42:22 ON 16 JUL 2004)

     FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS,
     LIFESCI' ENTERED AT 14:43:13 ON 16 JUL 2004
L1          1044 S "PROTEIN KINASE N" OR "PKN"
L2           226 S HUMAN AND L1
L3       6609293 S CLON? OR EXPRESS? OR RECOMBINANT
L4           141 S L2 AND L3
L5          1645 S RETINOBLASTOMAS OR PLACENTA(A)CHORIOCARCINOMAS OR BOCIO (A)TU
L6         66422 S LEUKEMIAS OR (WILM (2W) TUMOR?) OR "BRAIN (A) ANAPLASTIC(A)OL
L7         67972 S L5 OR L6
L8             2 S L4 AND L7
L9             2 DUP REM L8 (0 DUPLICATES REMOVED)
L10           68 DUP REM L4 (73 DUPLICATES REMOVED)
                E RUSCH D/AU
L11          161 S E3
                E KETCHUM K A/AU
L12          453 S E3-E7
                E DIFRANCESCO V/AU
L13          112 S E3-E4
L14          298 S BEASLEY E M/AU
L15          897 S L11 OR L12 OR L13 OR L14
L16            5 S L2 AND L15
L17            5 DUP REM L16 (0 DUPLICATES REMOVED)
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 NEWS  1           Web Page URLs for STN Seminar Schedule - N. America
 NEWS  2           "Ask CAS" for self-help around the clock
 NEWS  3  May 12   EXTEND option available in structure searching
 NEWS  4  May 12   Polymer links for the POLYLINK command completed in REGISTRY
 NEWS  5  May 27   New UPM (Update Code Maximum) field for more efficient patent
                   SDIs in CAplus
 NEWS  6  May 27   CAplus super roles and document types searchable in REGISTRY
 NEWS  7  Jun 22   STN Patent Forums to be held July 19-22, 2004
 NEWS  8  Jun 28   Additional enzyme-catalyzed reactions added to CASREACT
 NEWS  9  Jun 28   ANTE, AQUALINE, BIOENG, CIVILENG, ENVIROENG, MECHENG,
                   and WATER from CSA now available on STN(R)
 NEWS 10  Jul 12   BEILSTEIN enhanced with new display and select options,
                   resulting in a closer connection to BABS

 NEWS EXPRESS  MARCH 31 CURRENT WINDOWS VERSION IS V7.00A, CURRENT
               MACINTOSH VERSION IS V6.0c(ENG) AND V6.0Jc(JP),
               AND CURRENT DISCOVER FILE IS DATED 26 APRIL 2004
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accessing the remaining file names entered.
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                                                       ENTRY    SESSION
FULL ESTIMATED COST                                     0.42       0.42

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FILE 'LIFESCI' ENTERED AT 14:43:13 ON 16 JUL 2004
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=> s "protein kinase N" or "PKN"
L1         1044 "PROTEIN KINASE N" OR "PKN"

=> s human and l1
L2          226 HUMAN AND L1

=> s clon? or express? or recombinant
   5 FILES SEARCHED...
L3      6609293 CLON? OR EXPRESS? OR RECOMBINANT

=> s l2 and l3
L4          141 L2 AND L3

=> s retinoblastomas or placenta(a)choriocarcinomas or bocio (a)tumor?
L5         1645 RETINOBLASTOMAS OR PLACENTA(A) CHORIOCARCINOMAS OR BOCIO (A)
                TUMOR?

=> s leukemias or (Wilm (2w) tumor?) or "brain (a) anaplastic(a)oligodendromas"
   7 FILES SEARCHED...
L6        66422 LEUKEMIAS OR (WILM (2W) TUMOR?) OR "BRAIN (A) ANAPLASTIC(A)OLIGO
                DENDROMAS"

=> s l5 or l6
L7        67972 L5 OR L6

=> s l4 and l7
L8            2 L4 AND L7

=> dup rem l8
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PROCESSING COMPLETED FOR L8
L9 2 DUP REM L8 (0 DUPLICATES REMOVED)

=> d 1-2 ibib ab

L9 ANSWER 1 OF 2 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:594987 HCAPLUS
DOCUMENT NUMBER: 137:151129
TITLE: Protein, gene and cDNA sequences of a novel **human** protein kinase related to protein kinase **PKN** subfamily and their uses in drug screening
INVENTOR(S): Rusch, Douglas; Ketchum, Karen A.; Di Francesco, Valentina; Beasley, Ellen M.
PATENT ASSIGNEE(S): PE Corporation, USA
SOURCE: PCT Int. Appl., 76 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002061062	A2	20020808	WO 2002-US2152	20020129
WO 2002061062	A3	20030522		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
US 6500655	B1	20021231	US 2001-849334	20010507
EP 1358338	A2	20031105	EP 2002-713461	20020129
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				

PRIORITY APPLN. INFO.:
US 2001-773371 A 20010201
US 2001-849334 A 20010507
WO 2002-US2152 W 20020129

AB The invention provides protein, cDNA and genomic sequences for a novel **human** protein kinase related to protein kinase **PKN** subfamily. The protein kinase gene is **expressed** in **human** eye **retinoblastomas**, **placenta** **choriocarcinomas**, germ cells, **bocio tumors**, pre-B cell acute lymphoblastic **leukemias**, **wilm's** **tumors** of the kidney, uterus tumors, brain anaplastic oligodendromas, uterus endometrial adenocarcinomas, and leukocytes. The protein kinase gene has been mapped to chromosome 8. The invention also relates to screening modulator of said protein kinase and use them in therapy. The invention further relates to methods, vector and hosts for **expression** of said protein kinase.

L9 ANSWER 2 OF 2 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2003:1236 HCAPLUS
DOCUMENT NUMBER: 138:68934
TITLE: Identification, genomic and cDNA sequences and **cloning** of a **human protein** **kinase N** sequence homolog
INVENTOR(S): Rusch, Douglas; Ketchum, Karen A.; Di Francesco, Valentina; Beasley, Ellen M.
PATENT ASSIGNEE(S): Applera Corporation, USA

SOURCE: U.S., 44 pp., Cont.-in-part of U. S. Ser. No. 773,371, abandoned.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6500655	B1	20021231	US 2001-849334	20010507
WO 2002061062	A2	20020808	WO 2002-US2152	20020129
WO 2002061062	A3	20030522		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
EP 1358338	A2	20031105	EP 2002-713461	20020129
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
WO 2002090525	A2	20021114	WO 2002-US7155	20020308
WO 2002090525	A3	20030327		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
EP 1385863	A2	20040204	EP 2002-725095	20020308
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
US 2003049792	A1	20030313	US 2002-274878	20021022
US 6670163	B2	20031230		
US 2004067522	A1	20040408	US 2003-697266	20031031

PRIORITY APPLN. INFO.:
US 2001-773371 B2 20010201
US 2001-849334 A 20010507
WO 2002-US2152 W 20020129
WO 2002-US7155 W 20020308
US 2002-274878 A3 20021022

AB The present invention is based in part on the identification of amino acid sequences of **human** kinase peptides and proteins that are related to the **protein kinase N (PKN)** subfamily, as well as allelic variants and other mammalian orthologs thereof. The present invention provides genomic, cDNA and amino acid sequences of the **human protein kinase N** sequence homolog. Chromosomal mapping of the **protein kinase N** sequence homolog gene, tissue-specific **expression** profiles, and structural motifs of the polypeptides are provided. The protein and nucleic acid sequences of the invention, can be used as models for the development of **human** therapeutic targets, aid in the identification of therapeutic proteins, and serve as targets for the development of **human** therapeutic agents that modulate kinase activity in cells and tissues that **express** the kinase. **Expression** of the **protein kinase N** sequence homolog gene in **humans** in eye **retinoblastomas**

, **placenta choriocarcinomas**, germ cells, **bocio tumors**, pre-B cell acute lymphoblastic **leukemias**, **Wilm's tumors** of the kidney, uterus tumors, brain anaplastic oligodendromas, uterus endometrial adenocarcinomas, and leukocytes is reported.
REFERENCE COUNT: 1 THERE ARE 1 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

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(FILE 'HOME' ENTERED AT 14:42:22 ON 16 JUL 2004)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 14:43:13 ON 16 JUL 2004
L1 1044 S "PROTEIN KINASE N" OR "PKN"
L2 226 S HUMAN AND L1
L3 6609293 S CLON? OR EXPRESS? OR RECOMBINANT
L4 141 S L2 AND L3
L5 1645 S RETINOBLASTOMAS OR PLACENTA(A)CHORIOCARCINOMAS OR BOCIO (A)TU
L6 66422 S LEUKEMIAS OR (WILM (2W) TUMOR?) OR "BRAIN (A) ANAPLASTIC(A)OL
L7 67972 S L5 OR L6
L8 2 S L4 AND L7
L9 2 DUP REM L8 (0 DUPLICATES REMOVED)

=> dup rem l4
PROCESSING COMPLETED FOR L4
L10 68 DUP REM L4 (73 DUPLICATES REMOVED)

=> d 1-68 ibib ab

L10 ANSWER 1 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2004:203693 HCAPLUS
DOCUMENT NUMBER: 140:229429
TITLE: Protein and cDNA sequences of a **human protein kinase N** beta and use for treating cancer
INVENTOR(S): Klippel-Giese, Anke; Kaufmann, Joerg
PATENT ASSIGNEE(S): Atugen A.-G., Germany
SOURCE: PCT Int. Appl., 88 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2004019973	A1	20040311	WO 2003-EP8876	20030810
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
EP 1393742	A1	20040303	EP 2002-18572	20020814
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, SK				

PRIORITY APPLN. INFO.: EP 2002-18572 A 20020814

US 2002-409570P P 20020911

AB The present invention is related to use of **protein kinase N** beta or a fragment or derivative thereof as a downstream target of the PI 3-kinase pathway, preferably as a downstream drug target of the PI 3-kinase pathway. The present invention provides protein and cDNA sequences of a **human protein kinase N** beta. The present invention also is related to use of **protein kinase N** beta or a fragment or derivative thereof as a downstream target of the PI 3-kinase pathway, preferably as a downstream drug target of the PI 3-kinase pathway.

REFERENCE COUNT: 2 THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 2 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2004:178003 HCAPLUS
DOCUMENT NUMBER: 140:229419
TITLE: Protein and cDNA sequences of a **human protein kinase N** beta and use for treating cancer
INVENTOR(S): Klippel, Anke; Kaufmann, Joerg
PATENT ASSIGNEE(S): Atugen AG, Germany
SOURCE: Eur. Pat. Appl., 40 pp.
CODEN: EPXXDW
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 1393742	A1	20040303	EP 2002-18572	20020814
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, SK				
WO 2004019973	A1	20040311	WO 2003-EP8876	20030810
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
US 2004106569	A1	20040603	US 2003-640274	20030814

PRIORITY APPLN. INFO.: EP 2002-18572 A 20020814
US 2002-409570P P 20020911

AB The present invention provides protein and cDNA sequences of a **human protein kinase N** beta. The present invention also is related to use of **protein kinase N** beta or a fragment or derivative thereof as a downstream target of the PI 3-kinase pathway, preferably as a downstream drug target of the PI 3-kinase pathway.

REFERENCE COUNT: 2 THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 3 OF 68 MEDLINE on STN
ACCESSION NUMBER: 2004077549 MEDLINE
DOCUMENT NUMBER: PubMed ID: 14660612
TITLE: Structural insights into the interaction of ROCKI with the switch regions of RhoA.
AUTHOR: Dvorsky Radovan; Blumenstein Lars; Vetter Ingrid R; Ahmadian Mohammad Reza

CORPORATE SOURCE: Max-Planck-Institute fuer Molekulare Physiologie, Abteilung Strukturelle Biologie, Otto-Hahn-Strasse 11, 44227 Dortmund, Germany.
SOURCE: Journal of biological chemistry, (2004 Feb 20) 279 (8) 7098-104.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: PDB-1S1C
ENTRY MONTH: 200404
ENTRY DATE: Entered STN: 20040218
Last Updated on STN: 20040501
Entered Medline: 20040430

AB The Rho-ROCK pathway modulates the phosphorylation level of a variety of important signaling proteins and is thereby involved in miscellaneous cellular processes including cell migration, neurite outgrowth, and smooth muscle contraction. The observation of the involvement of the Rho-ROCK pathway in tumor invasion and in diseases such as hypertension and bronchial asthma makes it an interesting target for drug development. We herein present the crystal structure of the complex between active RhoA and the Rho-binding domain of ROCKI. The Rho-binding domain structure forms a parallel alpha-helical coiled-coil dimer and, in contrast to the published Rho-**protein kinase N** structure,
binds exclusively to the switch I and II regions of the guanosine 5'-(beta,gamma-imido)triphosphate-bound RhoA. The switch regions of two different RhoA molecules form a predominantly hydrophobic patch, which is complementarily bound by two identical short helices of 13 residues (amino acids 998-1010). The identified ROCK-binding site of RhoA strikingly supports the assumption of a common consensus-binding site for effector recognition.

L10 ANSWER 4 OF 68 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 2003368903 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12857884
TITLE: ROCK and nuclear factor-kappaB-dependent activation of cyclooxygenase-2 by Rho GTPases: effects on tumor growth and therapeutic consequences.
AUTHOR: Benitah Salvador Aznar; Valeron Pilar F; Lacal Juan Carlos
CORPORATE SOURCE: Department of Molecular and Cellular Biology of Cancer, Instituto de Investigaciones Biomedicas, Consejo Superior de Investigaciones Cientificas, Madrid, Spain.
SOURCE: Molecular biology of the cell, (2003 Jul) 14 (7) 3041-54.
Journal code: 9201390. ISSN: 1059-1524.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200404
ENTRY DATE: Entered STN: 20030808
Last Updated on STN: 20040414
Entered Medline: 20040413

AB Rho GTPases are overexpressed in a variety of **human** tumors contributing to both tumor proliferation and metastasis. Recently, several studies demonstrate an essential role of transcriptional regulation in Rho GTPases-induced oncogenesis. Herein, we demonstrate that RhoA, Rac1, and Cdc42 promote the **expression** of cyclooxygenase-2 (COX-2) at the transcriptional level by a mechanism that is dependent on the transcription factor nuclear factor-kappaB (NF-kappaB), but not Stat3, a transcription factor required for RhoA-induced tumorigenesis. With respect to RhoA, this effect is dependent on ROCK, but not **PKN**. Treatment of RhoA-, Rac1-, and Cdc42-transformed epithelial cells with Sulindac and NS-398, two

well-characterized nonsteroid antiinflammatory drugs (NSAIDs), results in growth inhibition as determined by cell proliferation assays. Accordingly, tumor growth of RhoA-**expressing** epithelial cells in syngeneic mice is strongly inhibited by NS-398 treatment. The effect of NSAIDs over RhoA-induced tumor growth is not exclusively dependent on COX-2 because DNA-binding of NF-kappaB is also abolished upon NSAIDs treatment, resulting in complete loss of COX-2 **expression**. Finally, treatment of RhoA-transformed cells with Bay11-7083, a specific NF-kappaB inhibitor, leads to inhibition of cell proliferation. We suggest that treatment of **human** tumors that overexpress Rho GTPases with NSAIDs and drugs that target NF-kappaB could constitute a valid antitumoral strategy.

L10 ANSWER 5 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2003:132181 HCAPLUS
DOCUMENT NUMBER: 138:381289
TITLE: Purification and crystallization of the N-terminal domain from the **human** doublecortin-like kinase
AUTHOR(S): Kim, Myung Hee; Derewenda, Urszula; Devedjiev, Yancho; Dauter, Zbigniew; Derewenda, Zygmunt S.
CORPORATE SOURCE: Department of Molecular Physiology and Biological Physics and the Cancer Center, University of Virginia, Charlottesville, VA, 22908-0736, USA
SOURCE: Acta Crystallographica, Section D: Biological Crystallography (2003), D59(3), 502-505
CODEN: ABCRE6; ISSN: 0907-4449
PUBLISHER: Blackwell Munksgaard
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The unique doublecortin-like tandem of 2 homologous domains is found in certain microtubule-associated proteins such as doublecortin (DCX) and doublecortin-like kinase (DCLK). It is responsible for interactions with tubulin/microtubules and regulates microtubule dynamics. Here, the **expression** and purification of the tandem from **human** DCLK (residues 49-280) and of the isolated domains (residues 49-154 and 176-280) and the successful crystallization of the N-terminal domain (N-DCLK) are reported. High-quality wild-type crystals were obtained and a complete native data set was collected to 1.5 Å resolution The crystals belonged to space group C2, with unit-cell parameters a = 85.98, b = 29.62, c = 40.33 Å, and β = 101.3°. Crystals of SeMet-substituted N-DCLK (L120M) were also obtained, but they exhibited the symmetry of space group P21, with unit-cell parameters a = 38.81, b = 29.43, c = 40.1 Å, and β = 115.7°.
REFERENCE COUNT: 18 THERE ARE 18 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 6 OF 68 MEDLINE on STN
ACCESSION NUMBER: 2003043428 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12514133
TITLE: A novel inducible transactivation domain in the androgen receptor: implications for PRK in prostate cancer.
AUTHOR: Metzger Eric; Muller Judith M; Ferrari Stefano; Buettner Reinhard; Schule Roland
CORPORATE SOURCE: Universitats-Frauenklinik und Zentrum fur Klinische Forschung, Klinikum der Universitat Freiburg, Breisacherstrasse 66, D-79106 Freiburg, Germany.
SOURCE: EMBO journal, (2003 Jan 15) 22 (2) 270-80.
Journal code: 8208664. ISSN: 0261-4189.
PUB. COUNTRY: England: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals

ENTRY MONTH: 200302
ENTRY DATE: Entered STN: 20030130
Last Updated on STN: 20030226
Entered Medline: 20030225

AB In addition to the classical activation by ligands, nuclear receptor activity is also regulated by ligand-independent signalling. Here, we unravel a novel signal transduction pathway that links the RhoA effector protein kinase C-related kinase PRK1 to the transcriptional activation of the androgen receptor (AR). Stimulation of the PRK signalling cascade results in a ligand-dependent superactivation of AR. We show that AR and PRK1 interact both in vivo and in vitro. The transactivation unit 5 (TAU-5) located in the N-terminus of AR suffices for activation by PRK1. Thus, TAU-5 defines a novel, signal-inducible transactivation domain. Furthermore, PRK1 promotes a functional complex of AR with the co-activator TIF-2. Importantly, PRK signalling also stimulates AR activity in the presence of adrenal androgens, which are still present in prostate tumour patients subjected to testicular androgen ablation therapy. Moreover, PRK1 activates AR even in the presence of the AR antagonist cyproterone acetate that is used in the clinical management of prostate cancer. Since prostate tumours strongly overexpress PRK1, our data support a model in which AR activity is controlled by PRK signalling.

L10 ANSWER 7 OF 68 MEDLINE on STN DUPLICATE 2
ACCESSION NUMBER: 2003239179 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12761180
TITLE: Regulation of a mitogen-activated protein kinase kinase kinase, MLTK by **PKN**.
AUTHOR: Takahashi Mikiko; Gotoh Yusuke; Isagawa Takayuki; Nishimura Tamako; Goyama Emiko; Kim Hon-Song; Mukai Hideyuki; Ono Yoshitaka
CORPORATE SOURCE: Biosignal Research Center and Graduate School of Science and Technology, Kobe University, Kobe 657-8501, Japan.
SOURCE: Journal of biochemistry, (2003 Feb) 133 (2) 181-7.
Journal code: 0376600. ISSN: 0021-924X.
PUB. COUNTRY: Japan
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200402
ENTRY DATE: Entered STN: 20030523
Last Updated on STN: 20040224
Entered Medline: 20040223

AB PKNalpha is a fatty acid- and Rho-activated serine/threonine protein kinase having a catalytic domain homologous to members of the protein kinase C family. Recently it was reported that PKNalpha is involved in the p38 mitogen-activated protein kinase (MAPK) signaling pathway. To date, however, how PKNalpha regulates the p38gamma MAPK signaling pathway is unclear. Here we demonstrate that PKNalpha efficiently phosphorylates MLTKalpha (MLK-like mitogen-activated protein triple kinase), which was recently identified as a MAPK kinase kinase (MAPKKK) for the p38 MAPK cascade. Phosphorylation of MLTKalpha by PKNalpha enhances its kinase activity in vitro. **Expression** of the kinase-negative mutant of PKNalpha inhibited the mobility shift of MLTKalpha caused by osmotic shock in SDS-PAGE. Furthermore, PKNalpha associates with each member of the p38gamma MAPK signaling pathway (p38gamma, MKK6, and MLTKalpha). These results suggest that PKNalpha functions as not only an upstream activator of MLTKalpha but also a putative scaffold protein for the p38gamma MAPK signaling pathway.

L10 ANSWER 8 OF 68 MEDLINE on STN DUPLICATE 3
ACCESSION NUMBER: 2003037299 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12545221
TITLE: **Cloning** and sequence analysis of tumor-associated gene hMMTAG2 from **human** multiple myeloma cell

line ARH-77.
AUTHOR: Tian Jing-Yan; Hu Wei-Xin; Tian Er-Ming; Shi Yi-Wu; Shen Qun-Xi; Tang Li-Jun; Jiang Yuan-Shan
CORPORATE SOURCE: Molecular Biology Research Center, Xiangya Medical College, Central South University, Changsha 410078, China.
SOURCE: Sheng wu hua xue yu sheng wu wu li xue bao Acta biochimica et biophysica Sinica, (2003 Feb) 35 (2) 143-8.
Journal code: 20730160R. ISSN: 0582-9879.
PUB. COUNTRY: China
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-AY137773
ENTRY MONTH: 200309
ENTRY DATE: Entered STN: 20030125
Last Updated on STN: 20030923
Entered Medline: 20030922

AB In order to look for the tumor-associated genes from **human** multiple myeloma (MM), a cDNA library of **human** multiple myeloma cell line ARH-77 was constructed with eukaryote **expression** vector pcDNA3.1(+). The length of inserted fragments in library was 1.2 kb in average. All **clones** in cDNA library were transferred in situ to nylon membrane, which was divided into eight equal parts (A-H) and cultured in LB medium to set up gene pools. The plasmids in cDNA library and in gene pools were extracted and NIH/3T3 cells were transfected respectively. By G418 screening and colonies counting, gene pool A was chosen for the second cycle transfection. After several cycles, a **clone**, A62-17, was obtained, which had significant transforming ability. The length of this **clone** was 993 bp. The RACE technique was used for rapid amplification of A62-17 5'-end. The full length of this sequence has 1300 bp and was named as hMMTAG2 gene. hMMTAG2 consists of 8 exons and codes for a polypeptide of 263 amino acids (the accession number in GenBank: AY137773). It was located at chromosome 1q42.13. hMMTAG2 had same transforming activities in NIH/3T3 cells as the **clone** A62-17, and the number of transformant foci was 6 folds more than the blank vector pcDNA3.1(+). The analysis of bioinformatics revealed that hMMTAG2 had many phosphorylation sites for several **protein kinases**, **N**-myristoylation sites and nuclear localization signals, so it may be a signal molecule in the nucleus.

L10 ANSWER 9 OF 68 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2004:202363 BIOSIS
DOCUMENT NUMBER: PREV200400202906
TITLE: Immunocytochemical study for 3 - phosphoinositide - dependent protein kinase 1 in Alzheimer brain tissues.
AUTHOR(S): Kawamata, T. [Reprint Author]; Mukai, H.; Takahashi, M.; Maeda, K.; Ono, Y.
CORPORATE SOURCE: Dept. Hlth. Sci, Kobe Univ. Sch. of Med, Kobe Univ, Kobe, Japan
SOURCE: Society for Neuroscience Abstract Viewer and Itinerary Planner, (2003) Vol. 2003, pp. Abstract No. 628.23.
http://sfn.scholarone.com. e-file.
Meeting Info.: 33rd Annual Meeting of the Society of Neuroscience. New Orleans, LA, USA. November 08-12, 2003. Society of Neuroscience.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 14 Apr 2004
Last Updated on STN: 14 Apr 2004

AB Neurofibrillary tangles (NFTs), one of pathological hallmarks in Alzheimer disease (AD), are composed of straight or paired helical filaments consisting of aberrantly hyperphosphorylated form of a

microtubule-associated protein tau. A serine/threonine kinase **PKN** directly phosphorylates tau in microtubule binding domains and triggers disruption of the microtubule array both in vitro and in vivo, and indirectly reduces the proline-directed tau phosphorylations recognized by the phosphorylation-dependent AT8, AT180 and AT270 antibodies. Here we report our findings on the **expression** of a protein kinase, 3-phosphoinositide-dependent protein kinase 1 (PDK1), which is known to phosphorylate and activate **PKN**, in AD brain tissues using immunocytochemistry. PDK1 was accumulated in a cluster of vesicles localized in the proximal dendrites and the cell bodies of neurons or in the glial filaments of some astrocytes in control **human** brains, although PDK1 is considered to be mainly a cytosolic protein. In AD damaged neurons, these vesicles were redistributed to intracellular NFTs and associating cytoplasmic granules, and were also relocated to such neuritic pathology as neuropil threads and degenerating neuritis within senile plaques, while extracellular NFTs were not immunolabeled for PDK1. Glial fibers were densely stained especially in the reactive astrocytes surrounding senile plaques in AD. Some of the neuronal vesicles were doubly labeled for PDK1 and **PKN**. Thus, PDK1 may work in the upstream of **PKN** and may be associated with the phosphorylation of tau leading to the formation of intracellular NFTs as well as the phosphorylation of glial fibrillary acidic protein in reactive astrocytes in AD brains.

L10 ANSWER 10 OF 68 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
DUPLICATE 4
ACCESSION NUMBER: 2002-11145 BIOTECHDS
TITLE: Peptide mimetic of a cytokine molecule, useful as a pharmaceutical, and in the manufacture of a medicament for the treatment or prevention of a disease in mammals, has atypical helix-turn-helix;
vector-mediated gene transfer and **expression** in Escherichia coli and transgenic animal model construction for use in peptidomics and allergy prevention and therapy
AUTHOR: SERRANO L; DOMINGUES H M
PATENT ASSIGNEE: EURO MOLECULAR BIOLOGY LAB
PATENT INFO: WO 2002012337 14 Feb 2002
APPLICATION INFO: WO 2000-IB1705 9 Aug 2000
PRIORITY INFO: GB 2000-19638 9 Aug 2000
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-227142 [28]
AB DERWENT ABSTRACT:
NOVELTY - A peptide mimetic (I) of a cytokine molecule comprising an atypical helix-turn-helix motif (II) mutated to incorporate one or more amino acid residues from the active site of the cytokine molecule, is new. (I) comprises a sequence of solup 10 comprising 45 amino acids fully defined in the specification.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following: (1) a nucleic acid molecule (III) encoding (I); (2) a vector (IV) comprising (III); (3) a host cell containing (III) or (IV); (4) a pharmaceutical composition (PC) comprising (I); (5) a diagnostic kit comprising (I); (6) a transgenic non-**human** mammal carrying a transgene encoding (I); (7) generating (I); (8) use of (II) as a template for the design of a peptide mimetic of a cytokine; and (9) preparing a cytokine receptor, by passing a composition containing the cytokine receptor over a matrix to which a peptide mimetic is bound.

WIDER DISCLOSURE - Also disclosed as new is a process for producing a transgenic non-**human** mammal carrying a transgene encoding (I).

BIOTECHNOLOGY - Preparation: (I) is generated, by incorporating the binding site of cytokine molecule into the sequence of an atypical helix-turn-helix motif, mutating the sequence of the generated peptide mimetic and selecting for variants of the sequence with improved

biological activity as a mimetic of a cytokine molecule (claimed). Preferred Protein: (II) is derived from the ROP protein (GenBank accession number P03051), the dimerization domain of Escherichia coli gene regulatory protein Arac (pdb code 2ara and 2aac) the ACC finger domain of the effector domain of protein kinase **PKN**/PRK1, or the coiled-coil of Thermus thermophilus seryl-tRNA synthetase (pdb code 1ser) Biou et al., 1994. (I) comprises a ROP helix-turn-helix monomer. The residues from the N terminus and/or the C terminus of (II) are deleted. The peptide sequence includes a Met residue at its N terminus. The cytokine molecule is a four helix bundle cytokine e.g. **human** growth hormone (HGH), granulocyte macrophage-colony stimulating factor (GM-CSF), granulocyte-colony stimulating factor (G-CSF), leukemia inhibitory factor (LIF), erythropoietin (EPO), interleukin (IL)-2 to IL-3, IL-4, IL-5, IL-6, IL-7, IL-9, IL-13, ciliary neurotrophic factor (CNTF), oncostatin (OSM) or an interferon. (I) binds to the IL-4 receptor, IL-2 receptor or HGH receptor with an affinity of at least 50 microM. The cytokine is IL-4, and cytokine receptor is IL-4Ralpha. (II) mutated to incorporate the active site of the cytokine molecule includes one or more substitutions such as Met 11 Ile, Ile 15 Glu, Glu 33 Lys, Ile 37 Arg, Ser 40 Lys, Leu 41 Arg , His 44 Arg , Ala 45 Asn and Glu 47 Trp.

ACTIVITY - Antiallergic. No supporting data given.

MECHANISM OF ACTION - None given.

USE - (I) is useful as a pharmaceutical, and in the manufacture of a medicament for the treatment or prevention of a disease in a mammal, preferably **human**. (I) or (II) is useful for preventing or treating a disease or condition in a patient. (I) is useful for preparing antibody against a cytokine, by immunizing an animal with (I) (claimed). (I) is useful for treating allergy-related conditions, and for the purification of target receptor, e.g. IL-4Ralpha.

ADVANTAGE - (I) is stable by virtue of structural features inherent in the atypical helix-turn-helix motif, yet incorporate amino acid residues from a cytokine molecule such that the mimetics bind to targets of the cytokine in question with a high affinity and high specificity.

EXAMPLE - ROP is an Escherichia coli transcription factor that regulates the copy number of ColE1 related plasmids. The protein sequence comprises 63 amino acids and the three-dimensional structure showed that they form a helix-turn-helix motif that dimerizes in solution. The functional protein contained two polypeptide chains that pack against each other and is very stable to temperature or chemically induced denaturation, with a Tm of 64 degrees C and a Cm (concentration of guanidium hydrochloride at the midpoint of the denaturation transition) of 3.3 M. The protein showed a typical coiled-coil fold. The interleukin (IL)-4 epitope was transferred to the surface of the ROP coiled-coil, at the dimer interface. The goal was to disrupt the hydrophobic interface in order to prevent the formation of dimers. The monomer thus obtained should recognize and bind IL-4Ralpha. The first and last seven terminal residues of the ROP sequence were deleted because in the best alignment obtained, they extended beyond the positions of interest. Ala 8 was replaced by Met to allow the over **expression** of protein in prokaryotic hosts. A Gly residue was introduced after Met to allow **cloning** into NcoI site. An N-terminal helix capping was designed by mutating Leu9 into Thr and Ala12 into Gln. Asn 10 was replaced by a negatively charged residue (Asp) to establish favorable interaction with helix macrodipole. The IL-4 binding site for IL-4Ralpha was introduced at the corresponding positions given in the specification. Mutations (replacement of Ser17, Thr21 and Asp43 by Ala) were designed in order to stabilize the ROP-derived IL-4 mimetic, and to increase the helical propensity of the sequence. In order to improve the packing of the two helices Cys38, His42, were replaced by Leu. Glu 39 was replaced by Arg in order to form a salt bridge with Asp36. The packing of the termini of the helices was improved by replacing the bulky side chain of Tyr49 by that of Phe. The helix was terminated by a Gly residue followed by a Ser. A 45-residue mini-protein designated as Solup10 was obtained. (43 pages)

L10 ANSWER 11 OF 68 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
ACCESSION NUMBER: 2003-00455 BIOTECHDS
TITLE: Novel immunoglobulin molecule for reducing tumor growth, binds to kinase insert domain-containing receptor with an affinity comparable to **human** vascular endothelial growth factor, and neutralizes activation of KDR; plasmid-mediated gene transfer for humanized antibody, chimeric antibody and single chain antibody production in COS cell for tumor therapy
AUTHOR: ZHU Z; WITTE L
PATENT ASSIGNEE: ZHU Z; WITTE L
PATENT INFO: US 2002064528 30 May 2002
APPLICATION INFO: US 2001-976787 12 Oct 2001
PRIORITY INFO: US 2001-976787 12 Oct 2001; US 2000-493539 28 Jan 2000
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-589175 [63]
AB DERWENT ABSTRACT:

NOVELTY - An immunoglobulin molecule (I) that binds to kinase insert domain-containing receptor (KDR) (a **human** homolog of mouse fetal liver kinase (FLK)-1 receptor) with an affinity comparable to **human** vascular endothelial growth factor (VEGF), and that neutralizes activation of KDR, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are included for the following: (1) a nucleic acid molecule (II) encoding: (a) a single chain antibody (Ia), a diabody (Ib), a triabody (Ic) or an antibody (Id) that neutralizes activation of KDR, comprising at least one variable heavy-chain fragment (F1) comprising CDRH1 (comprising a sequence GFNIKDFYMH), CDRH2 (comprising a sequence

WIDPENGDSGYAPKFQG), CDRH3 (comprising a sequence YYGDYEGY) or a sequence (S1) comprising 117 amino acids fully defined in the specification, and at least one variable light-chain fragment (F2) comprising CDRL1 (comprising a sequence SASSSVSYMH), CDRL2 (comprising a sequence STSNLAS, CDRL3 (comprising a sequence QQRSSYPFT) or a sequence (S2) comprising 108 amino acids fully defined in the specification; or (b) a peptide linker that covalently links F1 and F2; (2) a chimeric or humanized antibody (III) comprising (Id); and (3) making (I) (including (Ia), Ib), (Ic) or (Id)).

BIOTECHNOLOGY - Preparation: (I) (including (Ia), (Ib), (Ic) and (Id)) is obtained by inserting a nucleic acid molecule (II) into a host cell, and **expressing** the nucleic acid sequence (claimed).

Preferred Immunoglobulin: (I) comprises (Ia), (Ib), (Ic) or (Id). F1 (of (Ia)) and F2 (of (Ia)) are covalently linked by at least one peptide linker comprising at least 15 amino acids (a sequence (S3) GGGGSGGGGSGGGGS). F1 (of (Ib)) and F2 (of (Ib)) are covalently linked by a peptide linker comprising at least 5 and not more than 10 amino acids (comprising a sequence (S4) GGGGSGGGGS). (Ib) is monospecific or bispecific, and (Ic) is monospecific, dispecific or trispecific. (Ib) or (Ic) binds to at least one epitope on KDR.

ACTIVITY - Cytostatic.

MECHANISM OF ACTION - Neutralizes the activation of KDR; inhibitor of angiogenesis (claimed); inhibitor of VEGF-induced mitogenesis. The effect of anti-KDR antibodies on VEGF-stimulated mitogenesis of **human** endothelial cells was determined with a (3H)-TdR DNA incorporation assay using **human** umbilical vein endothelial cell (HUVEC). HUVEC (5x103 cells/well) were plated into 96-well tissue culture plates in 200 mul of EBM-2 medium without VEGF, basic fibroblast growth factor (bFGF) or epidermal growth factor (EGF) and incubated at 37degreesC for 72 hour. Various amounts of antibodies were added to duplicate wells and pre-incubated at 37degreesC for 1 hour, after which VEGF165 was added to a final concentration of 16 ng/ml. After 18 hours of incubation, 0.25 muCi of (3H)-TdR was added to each well and incubated for an additional 4 hours. DNA incorporated radioactivity was determined with a scintillation counter. Both c-p1C11 and scFv p1C11 effectively

inhibited mitogenesis of HUVEV stimulated VEGF. c-p1C11 was a stronger inhibitor of VEGF-induced mitogenesis of HUVEC than the parent scFv. The antibody concentrations required to inhibit 50% of VEGF-induced mitogenesis of HUVEC were 0.8 nM for c-p1C11 and 6 nM for the scFv, respectively. As expected, scFv p2A6 did not show any inhibitory effect on VEGF-stimulated endothelial cell proliferation.

USE - (I) (including (Ia), (Ib), (Ic) and (Id)) is useful for neutralizing the activation of KDR, reducing tumor growth and inhibiting angiogenesis (claimed).

EXAMPLE - The variable domains of the light (VL) and heavy (VH) chains of p1C11 were **cloned** from the scFv **expression** vector by polymerase chain reaction (PCR) using primer 1 (5'CTAGTAGCAACTGCAACTGGAGTACATTCAGACATCGAGCTC3') and primer 2 (5'TCGATCTAGAAGGATCCACTCACGTTTTATTTCCAG3'), and primer 3 (5'CTAGTAGCAACTGCAACTGGAGTACATTCACAGGTCAAGCTG3') and primer 4 (5'TCGAAGGATCCACTCACCTGAGGAGACGGT3'), respectively. The leader peptide sequence for protein secretion in mammalian cells was then added 5' to VL and VH by PCR using primer 2 and primer 5 (5'GGTCAAAAGCTTATGGGATGGTCATGTA TCATCCTTTTTCTAGTAGCAACT3'), and primers 5 and 4, respectively. Separate vectors for **expression** of chimeric IgG light chain and heavy chains were constructed. The **cloned** VL gene was digested with HindIII and BamHI, and ligated into the vector pKN100 containing the **human** kappa light chain constant region (CL) to create the **expression** vector for the chimeric p1C11 light chain, c-p1C11-L. The **cloned** VH gene was digested with HindIII and BamHI, and ligated into the vector pGID105 containing the **human** IgG1 (gamma) heavy chain constant domain (CH) to create the **expression** vector for the chimeric p1C11 heavy chain, c-p1C11-H. Both constructs were examined by restriction enzyme digestion and verified by dideoxynucleotide sequencing. Both the VH and the VL domains were precisely fused on their 5' ends to a gene segment encoding a leader peptide sequence as marked. The VH and the VL domains were ligated through HindIII/BamHI sites into **expression** vector pG1D105 containing a cDNA version of the **human** gamma1 constant region gene, and **pKN** 100 containing a cDNA version of the **human** kappa chain constant region gene, respectively. In each case, **expression** was under the control of the HCMVi promoter and terminated by an artificial termination sequence. COS cells were co-transfected with equal amounts of c-p1C11-L and c-p1C11-H plasmids for transient IgG **expression**. (34 pages)

L10 ANSWER 12 OF 68 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
ACCESSION NUMBER: 2002-17085 BIOTECHDS
TITLE: Identifying disruptors of biological pathways useful in therapeutic, diagnostic and related purposes, using genetic selection;
phosphorylation pathway disruptor drug screening for use as a therapeutic and in diagnosis
AUTHOR: MURRAY A W; SMITH D L; SORGER P K; NORMAN T C
PATENT ASSIGNEE: UNIV CALIFORNIA
PATENT INFO: US 6365347 2 Apr 2002
APPLICATION INFO: US 1997-58483 11 Apr 1997
PRIORITY INFO: US 1998-58483 10 Apr 1998
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-433422 [46]
AB DERWENT ABSTRACT:

NOVELTY - Identifying disruptors of biological pathways, comprising generating macromolecule libraries which are constructed by transforming host cells with a collection of recombination vectors that encode chimeras comprised of random peptide sequence and a carrier protein, and **expressing** the chimera intracellularly to identify peptide inhibitors of biological pathway through genetic selection, is new.

DETAILED DESCRIPTION - Identifying (M1) of a fusion macromolecule

(disruptors) which interacts within a cell with an intracellular target molecule (TM) and detecting if the interaction disrupts a biological pathway. The cell survives or proliferates when the interaction disrupts the pathway and the cell dies or fails to proliferate when the interaction does not disrupt the pathway. Desired macromolecule comprise a peptide library (PL) region which is presented in a restricted conformation by a heterologous carrier region, by: (a) generating a panel (P1) of nucleic acid sequences that encodes a peptide library, where P1 includes a nucleic acid sequence that encodes PL region that interacts with the intracellular TM within the cell; (b) inserting P1 into several vectors, where each vector comprises a carrier region to generate an **expression** library to chimeric vectors, where each chimeric vector has a nucleic acid sequence from P1 and a nucleic acid sequence encoding the carrier region having the restricted conformation; (c) introducing the **expression** library of chimeric vectors into several cells that lacks a recombination reporter gene so as to generate a library (L1) of cells; (d) growing L1 under suitable conditions to produce a library (L2) of fusion macromolecules within the cells, where L2 comprises PL region presented in a restricted conformation by the carrier region; and (e) selecting a desired cell that produces desired macromolecule from L2, by detecting the interaction between PL region and TM which results in disruption of the biological pathway within the desired cell and enables the desired cell to survive or proliferate, identifying the macromolecule produced by the cell so selected.

WIDER DISCLOSURE - (1) screening method which comprises growing the host cells under conditions which genetically select for **clones** that contain peptides that have the ability to disrupt the interactions between molecules that affects a biological process and isolating the vectors that encode the genetically selected peptides, by creating the affinity selection process one or more times, the plasmid encoding the desired peptides can be enriched, by increasing the stringency of the selection e.g. by decreasing the **expression** of the chimeras, increasing the temperature or varying other medium conditions peptides of increasingly higher affinity can be identified; (2) mutagenizing desired peptides to create inhibitors of varying affinities; and (3) identifying biological target a peptide inhibitor.

BIOTECHNOLOGY - Preferred Method: In M1, the interaction in step (e) between PL region and TM inhibits TM thereby disrupting the biological pathway within the desired cell, where the biological pathway is a phosphorylation pathway (Pw1) mediated by a cAMP-dependent kinase, yeast pheromone response pathway (Pw2), a cell cycle arrest pathway (Pw3), a cell DNA damage checkpoint pathway (Pw4), a cellular spindle assembly checkpoint pathway (Pw5) or a heat shock pathway (Pw6). The vectors of step (b) further comprises at least a promoter which is from bacterial promoter and yeast promoter, which is inducible such as GAL 1, HSP, CUP 1, PGK and Pho A, or a constitutive such as VP16 and ADH 1. The carrier region of the macromolecules comprises the nuclease loop of Staphyloccus aureus, and the target molecule is preferably target molecule for Pw2 is G- protein coupled receptor, for Pw3 is p34 molecule, for Pw4 is cdc5-ad molecule, and for Pw5 is an Mps1 molecule.

ACTIVITY - None given.

MECHANISM OF ACTION - Regulator of target protein activity (disruptors of biological pathways).

USE - M1 is useful for identifying disruptors of biological pathways (claimed), where the macromolecules thus identified are useful in therapeutic, diagnostic and related purposes.

ADVANTAGE - This screening does not require a detailed biochemical understanding of the pathway and can provide inhibitors of several different steps of the same pathway. This screening entails the step of growing the host cells under a specified set of conditions that genetically select for **clones** that contain peptides that have the ability to affect the interactions between biologically important molecules, by presenting a peptide that is constrained by its placement in the surface loop of a carrier protein, a library of peptides was

produced containing peptides that were more conformationally restricted and can interact with their binding partners with higher affinity than was observed for unconstrained peptides. The intracellular **expression** of the conformationally constrained peptide library provides advantages over the prior art in that the peptide library was a wide access to diverse number of intracellular targets. In this manner, the members of this library have the potential to interact with a wide variety of molecules, involved in growth and regulatory processes within the cell. Unlike the libraries disclosed in the prior art, the construction of this peptide library allows the desired sequences to be easily modified and manipulated. By repeating the affinity selection process one or more times, desired peptides could be enriched. By increasing the stringency of the selection e.g. by increasing the temperature or other medium conditions, peptides of increasing higher affinity can be identified. By manipulating the sequence of a desired peptide via the use of the disclosed procedures such as error prone polymerase chain reaction (PCR), variants with altered biological activity could be generated, isolated and characterized.

EXAMPLE - An oligonucleotide encoding a 22 amino acid peptide was synthesized which was reported to inhibit cAMP-dependent protein kinase with nM affinity. As a control, a non-inhibitory peptide chimera was also constructed. The oligonucleotide sequences encoding these peptides were constructed to incorporate Eco-RI and XhoI sequences to facilitate its unidirectional **cloning**. The oligonucleotide **clones** were ligated into the external loop of an engineered Staphylococcus aureus nuclease coding region of the gel purified pSF248 plasmid which was digested EcoRI-XhoI. The fusion protein (peptide chimera) backbone was engineered for easy use and included an HA tag, and a polyhistidine stretch. In yeast pSF248 **expressed** the peptide-nuclease chimera under the control of the GAL1 promoter. The above ligation mixture was transformed into bacterial strains SE6004 (r+m+N lacU169 araD139 relA strR thilamB560 prlA4) or TG1 and selected on ampicillin. Individual colonies were isolated, sequenced to verify the reading frame and plasmids were prepared. Plasmids containing the 22 amino acid peptide were transformed into the LL8 yeast strain (derived from SP1, Toda et al., (1987) Cell 50, 277-284 which **expressed human** cAMP-dependent protein kinase and deletions in three genes encoding catalytic subunits of yeast cAMP-dependent protein kinase (TPK1, TPK2, TPK3) and **expressed** the **human** cAMP-dependent protein kinase and selected from growth on medium lacking uracil. For optimal **expression** of the inducible peptide chimeras, yeast strains were grown in synthetic media with 2 % galactose and lacking uracil prepared. The genetic selection in this protocol entailed an analysis of growth on glucose and galactose-containing medium, where the growth of individual cells was quantified over time by counting the number of cells in cell colonies. This data was compared to data generated under identical conditions from a control peptide-nuclease chimera (**PKN** loop) that carried a mutant version of peptide that failed to inhibit cAMP-dependent protein kinase. The presence of the inhibitory peptide chimera (PKI loop) produced a marked difference in yeast growth patterns. Protein for in vitro studies was **expressed** in bacteria and purified. In this procedure, the pSF248-**PKN** plasmid was cut with the restriction endonuclease and BamHI and religated in order to allow efficient bacterial **expression** of the peptide chimera. The Bam-H1 cut and re-ligated plasmid was transformed into bacterial strain SE6004 and a chimeric protein preparation was generated via the periplasmic protein preparation methods. The peptide-nuclease chimeras from the periplasmic protein preparation were then purified by metal affinity chromatography. An in vitro analysis of the properties of the peptide chimera was undertaken where the biochemical inhibition of cAMP-dependent protein kinase was evaluated in vitro. The inhibitory effects of the chimeric peptides on the reaction were observed by monitoring the differental 32P incorporation into the BCY1 substrate by gel electrophoresis.(39 pages)

L10 ANSWER 13 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:615889 HCAPLUS
DOCUMENT NUMBER: 137:180730
TITLE: **Human** cDNA/DNA molecules and proteins encoded by them with enhanced **expression** in apoptosis-resistant cell **clones**, and use thereof in diagnosis, therapeutics and drug screening
INVENTOR(S): Ullrich, Axel; Abraham, Reimar
PATENT ASSIGNEE(S): Max-Planck-Gesellschaft zur Foerderung der Wissenschaften e.V., Germany
SOURCE: PCT Int. Appl., 56 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002063037	A2	20020815	WO 2002-EP1073	20020201
WO 2002063037	A3	20031002		
WO 2002063037	C2	20040219		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
EP 1364066	A2	20031126	EP 2002-718083	20020201
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
JP 2004517638	T2	20040617	JP 2002-562773	20020201
US 2004110177	A1	20040610	US 2003-470845	20030731

PRIORITY APPLN. INFO.: US 2001-265631P P 20010202
WO 2002-EP1073 W 20020201

AB The present invention relates to a method for identifying nucleic acid mols. functionally associated with a desired phenotype, such as cancer cell properties, including anti-apoptosis. The method, which allows for generation of **expression** profiles of genes associated with said desired phenotype, involves a mutagenesis and/or genome rearrangement step, followed by selection of cell **clones** displaying the desired phenotype. The invention also relates that the method involves the use of the following techniques: fluorescence-activated cell sorting (FACS); nucleic acid microarray (cDNA, genomic or oligonucleotide); protein array; two-dimensional gel electrophoresis; and/or mass spectrometry. The invention further relates that the disclosed method was used to identify genes, which are differentially **expressed** in apoptosis-sensitive and apoptosis-resistant cells. Specifically, the invention relates that apoptosis was induced in **human** cervix carcinoma cell line HeLa S3 by Fas activation. After the selection procedure, only a low number of living cells were present, which had a higher resistance against apoptosis than the parental cell line. MRNA was isolated from these surviving **clones**, and from the parental cell line, and transcribed into cDNA. CDNA microarray technol. was used to identify about 150-200 genes (cDNA/DNA mols.) that exhibited enhanced **expression** in apoptosis-resistant **clones**. The GenBank accession nos. of some of these cDNA/DNA mols. are provided, along with the products encoded by said mols. Still further, the invention relates that most of the apoptosis-associated genes encode protein phosphatases, and

kinases. Finally, the invention relates that said nucleic acid mols., and proteins encoded by mols., can be used as targets in diagnosis, therapeutics and drug screening, particularly for disorders associated with dysfunction of apoptotic processes, such as tumors.

L10 ANSWER 14 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:594987 HCAPLUS
DOCUMENT NUMBER: 137:151129
TITLE: Protein, gene and cDNA sequences of a novel **human** protein kinase related to protein kinase **PKN** subfamily and their uses in drug screening
INVENTOR(S): Rusch, Douglas; Ketchum, Karen A.; Di Francesco, Valentina; Beasley, Ellen M.
PATENT ASSIGNEE(S): PE Corporation, USA
SOURCE: PCT Int. Appl., 76 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002061062	A2	20020808	WO 2002-US2152	20020129
WO 2002061062	A3	20030522		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
US 6500655	B1	20021231	US 2001-849334	20010507
EP 1358338	A2	20031105	EP 2002-713461	20020129
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				

PRIORITY APPLN. INFO.:
US 2001-773371 A 20010201
US 2001-849334 A 20010507
WO 2002-US2152 W 20020129

AB The invention provides protein, cDNA and genomic sequences for a novel **human** protein kinase related to protein kinase **PKN** subfamily. The protein kinase gene is **expressed** in **human** eye retinoblastomas, placenta choriocarcinomas, germ cells, bocio tumors, pre-B cell acute lymphoblastic leukemias, wilm's tumors of the kidney, uterus tumors, brain anaplastic oligodendromas, uterus endometrial adenocarcinomas, and leukocytes. The protein kinase gene has been mapped to chromosome 8. The invention also relates to screening modulator of said protein kinase and use them in therapy. The invention further relates to methods, vector and hosts for **expression** of said protein kinase.

L10 ANSWER 15 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:409190 HCAPLUS
DOCUMENT NUMBER: 137:1566
TITLE: Protein, gene and cDNA sequences of a novel **human protein kinase N** sequence homolog
INVENTOR(S): Wei, Ming-hui; Chandramouliswaran, Ishwar; Ye, Jane; Ketchum, Karen A.; Di Francesco, Valentina; Beasley, Ellen M.
PATENT ASSIGNEE(S): USA

SOURCE: U.S. Pat. Appl. Publ., 39 pp., Cont.-in-part of U.S. Ser. No. 734,032.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2002064851	A1	20020530	US 2001-816094	20010326
US 6534299	B2	20030318		
US 2002103116	A1	20020801	US 2000-734032	20001212
WO 2001088148	A2	20011122	WO 2001-US15776	20010517
WO 2001088148	A3	20031016		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
EP 1373516	A2	20040102	EP 2001-952118	20010517
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
JP 2004507223	T2	20040311	JP 2001-584530	20010517
US 2003022339	A1	20030130	US 2002-233613	20020904
US 6649389	B2	20031118		
US 2004038362	A1	20040226	US 2003-639429	20030813

PRIORITY APPLN. INFO.:

US 2000-205228P	P	20000517
US 2000-734032	A2	20001212
US 2001-816094	A	20010326
WO 2001-US15776	W	20010517
US 2002-233613	A3	20020904

AB The invention provides protein, cDNA and genomic sequences for a novel **human** protein, which shares sequence homol. to a known kinase and is related to the **protein kinase N** subfamily. The kinase sequence homolog gene is **expressed** in **humans** in the brain, placenta, kidney and heart. Seven one novel single nucleotide polymorphism sites (beyond the ORF or in intron regions), including three indels, have been identified on kinase sequence homolog gene. Thus, the present invention specifically provides isolated protein and nucleic acid mols., methods of identifying orthologs and paralogs of the kinases, methods of identifying modulators of the kinases, and methods of diagnosis and treatment of diseases associated with the kinase.

L10 ANSWER 16 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2003:1236 HCAPLUS
DOCUMENT NUMBER: 138:68934
TITLE: Identification, genomic and cDNA sequences and **cloning** of a **human protein kinase N** sequence homolog
INVENTOR(S): Rusch, Douglas; Ketchum, Karen A.; Di Francesco, Valentina; Beasley, Ellen M.
PATENT ASSIGNEE(S): Applera Corporation, USA
SOURCE: U.S., 44 pp., Cont.-in-part of U. S. Ser. No. 773,371, abandoned.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6500655	B1	20021231	US 2001-849334	20010507
WO 2002061062	A2	20020808	WO 2002-US2152	20020129
WO 2002061062	A3	20030522		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
EP 1358338	A2	20031105	EP 2002-713461	20020129
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
WO 2002090525	A2	20021114	WO 2002-US7155	20020308
WO 2002090525	A3	20030327		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
EP 1385863	A2	20040204	EP 2002-725095	20020308
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
US 2003049792	A1	20030313	US 2002-274878	20021022
US 6670163	B2	20031230		
US 2004067522	A1	20040408	US 2003-697266	20031031

PRIORITY APPLN. INFO.:

US 2001-773371	B2	20010201
US 2001-849334	A	20010507
WO 2002-US2152	W	20020129
WO 2002-US7155	W	20020308
US 2002-274878	A3	20021022

AB The present invention is based in part on the identification of amino acid sequences of **human** kinase peptides and proteins that are related to the **protein kinase N** (**PKN**) subfamily, as well as allelic variants and other mammalian orthologs thereof. The present invention provides genomic, cDNA and amino acid sequences of the **human protein kinase N** sequence homolog. Chromosomal mapping of the **protein kinase N** sequence homolog gene, tissue-specific **expression** profiles, and structural motifs of the polypeptides are provided. The protein and nucleic acid sequences of the invention, can be used as models for the development of **human** therapeutic targets, aid in the identification of therapeutic proteins, and serve as targets for the development of **human** therapeutic agents that modulate kinase activity in cells and tissues that **express** the kinase. **Expression** of the **protein kinase N** sequence homolog gene in **humans** in eye retinoblastomas, placenta choriocarcinomas, germ cells, bocio tumors, pre-B cell acute lymphoblastic leukemias, Wilm's tumors of the kidney, uterus tumors, brain anaplastic oligodendromas, uterus endometrial adenocarcinomas, and leukocytes is reported.

REFERENCE COUNT: 1 THERE ARE 1 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 17 OF 68 MEDLINE on STN
ACCESSION NUMBER: 2002493302 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12119292
TITLE: cGMP-dependent protein kinase inhibits serum-response element-dependent transcription by inhibiting rho activation and functions.
AUTHOR: Gudi Tanima; Chen Jeffrey C; Casteel Darren E; Seasholtz Tammy M; Boss Gerry R; Pilz Renate B
CORPORATE SOURCE: Department of Medicine, University of California, San Diego, La Jolla, California 92093-0652, USA.
CONTRACT NUMBER: CA89828 (NCI)
GM55586 (NIGMS)
SOURCE: Journal of biological chemistry, (2002 Oct 4) 277 (40) 37382-93.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200211
ENTRY DATE: Entered STN: 20021001
Last Updated on STN: 20030105
Entered Medline: 20021120

AB RhoA, in its active GTP-bound form, stimulates transcription through activation of the serum-response factor (SRF). We found that cGMP inhibited serum-induced Rho.GTP loading and transcriptional activation of SRF-dependent reporter genes in smooth muscle and glial cells in a cGMP-dependent protein kinase (G-kinase)-dependent fashion. Serum stimulation of the SRF target gene vinculin was also blocked by cGMP/G-kinase. G-kinase activation inhibited SRF-dependent transcription induced by upstream RhoA activators including Galpha(13) and p115RhoGEF, with Galpha(13)-induced Rho.GTP loading inhibited by G-kinase. G-kinase had no effect on the high activation levels of RhoA(63L) or the double mutant RhoA(63L,188A) but inhibited transcriptional activation by these two RhoA mutants to a similar extent, suggesting an effect downstream of RhoA and independent of RhoA Ser(188) phosphorylation. Constitutively active forms of the Rho effectors Rho kinase (ROK), **PKN**, and PRK-2 induced SRF-dependent transcription in a cell type-specific fashion with ROK being the most efficient; G-kinase inhibited transcription induced by all three effectors without affecting ROK catalytic activity. G-kinase had no effect on RhoA(63L)-induced morphological changes in glial cells, suggesting distinct transcriptional and cytoskeletal effectors of RhoA. We conclude that G-kinase inhibits SRF-dependent transcription by interfering with RhoA signaling; G-kinase acts both upstream of RhoA, inhibiting serum- or Galpha(13)-induced Rho activation, and downstream of RhoA, inhibiting steps distal to the Rho targets ROK, **PKN**, and PRK-2.

L10 ANSWER 18 OF 68 MEDLINE on STN DUPLICATE 5
ACCESSION NUMBER: 2002697966 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12459919
TITLE: **Cloning** and characterisation of PKB and PRK homologs from Hydra and the evolution of the protein kinase family.
AUTHOR: Herold Michaela; Cikala Mihai; MacWilliams Harry; David Charles N; Bottger Angelika
CORPORATE SOURCE: Ludwig Maximilians-University Munich, Zoological Institute, Luisenstrasse 14, 80333 Munich, Germany.
SOURCE: Development genes and evolution, (2002 Dec) 212 (11) 513-9.
Journal code: 9613264. ISSN: 0949-944X.
PUB. COUNTRY: Germany: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English

FILE SEGMENT: Priority Journals
ENTRY MONTH: 200309
ENTRY DATE: Entered STN: 20021217
Last Updated on STN: 20030928
Entered Medline: 20030926

AB Two new serine/threonine protein kinases have been **cloned** from Hydra cDNA. The first of these kinases belongs to the PKB/Akt family. It is **expressed** ubiquitously in Hydra at a relatively low level but is upregulated during head regeneration. The second kinase is a member of the PRK/**PKN** family. It is ubiquitously **expressed** in Hydra tissue, albeit at a higher level than PKB. Construction of a phylogenetic tree including the Hydra PRK and PKB kinases and two PKC homologs previously **cloned** by Hassel and comparing them with members of the PKC, PKB and PRK families from porifera, Dictyostelium,yeast, Drosophila, Caenorhabditis and **humans** provide support for a simple model for the evolution of these kinase families. An ancestral precursor which contained a pleckstrin homology domain in its N-terminus and a C-terminal kinase domain gave rise to PKB in Dictyostelium. From this ancestor the PKB/PRK and PKC families evolved. The pleckstrin homology domain was lost in the PKC and PRK families and kept in the PKB family. PKB homologs have now been found in a variety of multicellular animals with Hydra being the phylogenetically earliest representative. Members of the PRK/PKC family, on the other hand, are also present in fungi. The precursor for these kinases must have contained N-terminal regulatory domains that were retained in fungal PRKs but subsequently partitioned between kinases of the PKC and PRK groups in metazoans.

L10 ANSWER 19 OF 68 EMBASE
on STN
ACCESSION NUMBER: 2002198699 EMBASE
TITLE: Smooth muscle cells on the move: The battle for actin.
AUTHOR: Doevendans P.A.; Van Eys G.
CORPORATE SOURCE: P.A. Doevendans, Department of Cardiology, Cardiovascular Research Institute, Academic Hospital Maastricht, 6202 AZ Maastricht, Netherlands. p.doevendans@cardio.azm.nl
SOURCE: Cardiovascular Research, (2002) 54/3 (499-502).
Refs: 25
ISSN: 0008-6363 CODEN: CVREAU
PUBLISHER IDENT.: S 0008-6363(02)00395-4
COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Editorial
FILE SEGMENT: 018 Cardiovascular Diseases and Cardiovascular Surgery
029 Clinical Biochemistry
LANGUAGE: English

L10 ANSWER 20 OF 68 MEDLINE on STN
ACCESSION NUMBER: 2002055825 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11777936
TITLE: Fyn tyrosine kinase is a downstream mediator of Rho/PRK2 function in keratinocyte cell-cell adhesion.
AUTHOR: Calautti Enzo; Grossi Maddalena; Mammucari Cristina; Aoyama Yumi; Pirro Maria; Ono Yoshitaka; Li Jie; Dotto G Paolo
CORPORATE SOURCE: Cutaneous Biology Research Center, Massachusetts General Hospital and Harvard Medical School, Charlestown, MA 02129.
CONTRACT NUMBER: AR39190 (NIAMS)
CA16038 (NCI)
CA73796 (NCI)
SOURCE: Journal of cell biology, (2002 Jan 7) 156 (1) 137-48.
Journal code: 0375356. ISSN: 0021-9525.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals

ENTRY MONTH: 200202
ENTRY DATE: Entered STN: 20020125
Last Updated on STN: 20030105
Entered Medline: 20020214

AB The Rho GTPase and Fyn tyrosine kinase have been implicated previously in positive control of keratinocyte cell-cell adhesion. Here, we show that Rho and Fyn operate along the same signaling pathway. Endogenous Rho activity increases in differentiating keratinocytes and is required for both Fyn kinase activation and increased tyrosine phosphorylation of beta- and gamma-catenin, which is associated with the establishment of keratinocyte cell-cell adhesion. Conversely, **expression** of constitutive active Rho is sufficient to promote cell-cell adhesion through a tyrosine kinase- and Fyn-dependent mechanism, trigger Fyn kinase activation, and induce tyrosine phosphorylation of beta- and gamma-catenin and p120ctn. The positive effects of activated Rho on cell-cell adhesion are not induced by an activated Rho mutant with defective binding to the serine/threonine PRK2/**PKN** kinases. Endogenous PRK2 kinase activity increases with keratinocyte differentiation, and, like activated Rho, increased PRK2 activity promotes keratinocyte cell-cell adhesion and induces tyrosine phosphorylation of beta- and gamma-catenin and Fyn kinase activation. Thus, these findings reveal a novel role of Fyn as a downstream mediator of Rho in control of keratinocyte cell-cell adhesion and implicate the PRK2 kinase, a direct Rho effector, as a link between Rho and Fyn activation.

L10 ANSWER 21 OF 68 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
ACCESSION NUMBER: 2002-07405 BIOTECHDS
TITLE: **Human** kinase protein and polynucleotides encoding them, useful for identifying modulators of kinase polypeptides and for treating, preventing, and/or diagnosing neurodegenerative diseases and cancer; vector-mediated **recombinant** protein gene transfer and **expression** in host cell, DNA probe, antibody, DNA chip and transgenic animal for disease prevention, diagnosis and gene therapy
AUTHOR: WEI M; CHANDRAMOULISWARA I; YE J; KETCHUM K A; DI FRANCESCO V; BEASLEY E M
PATENT ASSIGNEE: APPLERA CORP
PATENT INFO: WO 2001088148 22 Nov 2001
APPLICATION INFO: WO 2000-US15776 17 May 2000
PRIORITY INFO: US 2001-816094 26 Mar 2001
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-089857 [12]

AB DERWENT ABSTRACT:
NOVELTY - An isolated protein (a member of kinase family of protein and is related to **PKN** kinase subfamily) consisting or comprising a fully defined sequence of 424 amino acids (S2) as given in the specification, or its fragment comprising 10 contiguous amino acids, or an amino acid sequence of an allelic variant or ortholog of the amino acid sequence of (S2), is new.

DETAILED DESCRIPTION - An isolated protein (a member of kinase family of protein and is related to **PKN** kinase subfamily) consisting or comprising a fully defined sequence of 424 amino acids (S2) as given in the specification, or its fragment comprising 10 contiguous amino acids, or an amino acid sequence of an allelic variant or ortholog of the amino acid sequence of (S2), is new. (I) consists of or comprises: an amino acid sequence of (S2); an amino acid sequence of an allelic variant or an ortholog of (S2), where the allelic variant or ortholog is encoded by a nucleic acid molecule that hybridizes under stringent conditions to the opposite strand of a nucleic acid molecule having a fully defined sequence of 2598 nucleotides (S1) (transcript/cDNA) or 7301 nucleotides (S3) (genomic DNA) as given in the specification; a fragment of an amino acid sequence of (S2), comprising 10 contiguous amino acids.

INDEPENDENT CLAIMS are also included for the following: (1) an isolated antibody (II) that selectively binds to (I) comprising the amino acid sequence of (S2), its allelic variant or ortholog, or fragment; (2) an isolated nucleic acid molecule (III) consisting or comprising of a nucleotide sequence that encodes (I) or a nucleotide sequence that is complement of the nucleotide sequence encoding (I); (3) a gene chip comprising (III) that comprises a nucleotide sequence encoding (I), or its complement; (4) a transgenic non-**human** animal comprising (III) that comprises a nucleotide sequence encoding (I), or its complement; (5) a nucleic acid vector (IV) comprising (III) that comprises a nucleotide sequence encoding (I), or its complement; (6) a host cell comprising (IV); (7) preparation of (I); (8) detecting the presence of (I) comprising the amino acid sequence of (S2), its allelic variant or ortholog, or fragment, in a sample involves contacting the sample with a detection agent that specifically allows detection of the presence of the peptide in the sample and then detecting the presence of the peptide; (9) detecting the presence of (III) in a sample involves contacting the sample with an oligonucleotide that hybridizes to the nucleic acid molecule under stringent conditions and determining whether a oligonucleotide binds to the nucleic acid molecule in the sample; (10) a pharmaceutical composition (V) comprising an agent that binds to (I) comprising an amino acid sequence of (S2), its allelic variant or ortholog or fragment, and a carrier; (10) an isolated **human** kinase peptide (VI) having an amino acid sequence that shares 70% homology with (S2); and (11) an isolated nucleic acid molecule (VII) encoding a **human** kinase peptide which shares at least 80% homology with (S1) or (S3).

WIDER DISCLOSURE - The following are disclosed: (1) isolated peptide and protein molecules that consist essentially of the amino acid sequence of (S2), its allelic variant or ortholog, or fragment; (2) nucleic acid molecules that consist essentially of nucleotide sequence that encodes (I) or a nucleotide sequence that is complement of the nucleotide sequence encoding (I); (3) chimeric or fusion proteins comprising (I); (4) derivatives or analogs of (I) in which a substituted amino acid residue is not one encoded by the genetic code; (5) paralogs of the kinase polypeptide; (6) novel agents identified by the above mentioned screening methods;(7) kit comprising (II) for detecting (I) comprising an amino acid sequence of (S2), its allelic variant or ortholog or fragment; (8) non-coding fragments of a nucleic acid molecule having a sequence of (S1) or (S3); and (9) kits for detecting the presence of kinase protein nucleic acid in a biological sample.

BIOTECHNOLOGY - Preparation: (I) is prepared by standard **recombinant** techniques (claimed). Preferred Molecules: (VI) shares 90% homology with (S2), and (VII) shares at least 90% homology with (S1) or (S3).

ACTIVITY - Cytostatic; neuroprotective.

MECHANISM OF ACTION - Gene therapy; **human** kinase protein **expression** or activity modulator. No supporting data is given.

USE - The nucleic acids and polypeptides may be used in the prevention, diagnosis and treatment of diseases associated with inappropriate kinase **expression**. For example, the nucleic acids (or vectors containing them) and the kinase may be used to treat disorders associated with decreased **expression** by rectifying mutations or deletions in a patient's genome that affect the activity of the enzyme by **expressing** inactive proteins or to supplement the patients own production of kinases. Additionally, the nucleic acids may be used to produce the kinase, by inserting the nucleic acids into a host cell and culturing the cell to **express** the protein. The nucleic acid and its complementary sequences may also be used as DNA probes in diagnostic assays to detect and quantitate the presence similar nucleic acids in samples, and therefore which patients may be in need of restorative therapy. The polypeptides may also be used as antigens in the production of antibodies against the kinase and in assays to identify modulators of kinase **expression** and activity. The anti-kinase

antibodies and antagonists may also be used to down regulate **expression** and activity. The anti-kinase antibodies may also be used as diagnostic agents for detecting the presence of kinase polypeptides in samples (e.g. by enzyme linked immunosorbant assay (ELISA)). Disorders that may be prevented, diagnosed and/or treated by the above methods include, for example neurodegenerative diseases.
ADMINISTRATION - No specific administration details are given.
EXAMPLE - None given.(65 pages)

L10 ANSWER 22 OF 68 MEDLINE on STN DUPLICATE 6
ACCESSION NUMBER: 2001315011 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11259428
TITLE: **PKN** regulates phospholipase D1 through direct interaction.
AUTHOR: Oishi K; Takahashi M; Mukai H; Banno Y; Nakashima S; Kanaho Y; Nozawa Y; Ono Y
CORPORATE SOURCE: Graduate School of Science and Technology, and the Biosignal Research Center, Kobe University, Kobe 657-8501, Japan.
SOURCE: Journal of biological chemistry, (2001 May 25) 276 (21) 18096-101.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200107
ENTRY DATE: Entered STN: 20010709
Last Updated on STN: 20030105
Entered Medline: 20010705
AB The association of phospholipase (PLD)-1 with protein kinase C-related protein kinases, PKNalpha and PKNbeta, was analyzed. PLD1 interacted with PKNalpha and PKNbeta in COS-7 cells transiently transfected with PLD1 and PKNalpha or PKNbeta **expression** constructs. The interactions between endogenous PLD1 and PKNalpha or PKNbeta were confirmed by co-immunoprecipitation from mammalian cells. In vitro binding studies using the deletion mutants of PLD1 indicated that PKNalpha directly bound to residues 228-598 of PLD1 and that PKNbeta interacted with residues 1-228 and 228-598 of PLD1. PKNalpha stimulated the activity of PLD1 in the presence of phosphatidylinositol 4,5-bisphosphate in vitro, whereas PKNbeta had a modest effect on the stimulation of PLD1 activity. The stimulation of PLD1 activity by PKNalpha was slightly enhanced by the addition of arachidonic acid. These results suggest that the **PKN** family functions as a novel intracellular player of PLD1 signaling pathway.

L10 ANSWER 23 OF 68 MEDLINE on STN DUPLICATE 7
ACCESSION NUMBER: 2001169889 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11104762
TITLE: Phosphorylation of tau is regulated by **PKN**.
COMMENT: Erratum in: J Biol Chem 2001 Jun 22;276(25):23212
AUTHOR: Taniguchi T; Kawamata T; Mukai H; Hasegawa H; Isagawa T; Yasuda M; Hashimoto T; Terashima A; Nakai M; Mori H; Ono Y; Tanaka C
CORPORATE SOURCE: Hyogo Institute for Aging Brain and Cognitive Disorders, Himeji 670-0981, Japan.. tanigu@hiabcd.go.jp
SOURCE: Journal of biological chemistry, (2001 Mar 30) 276 (13) 10025-31.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200105

ENTRY DATE: Entered STN: 20010517
Last Updated on STN: 20030105
Entered Medline: 20010510

AB For the phosphorylation state of microtubule-associated protein, tau plays a pivotal role in regulating microtubule networks in neurons. Tau promotes the assembly and stabilization of microtubules. The potential for tau to bind to microtubules is down-regulated after local phosphorylation. When we investigated the effects of **PKN** activation on tau phosphorylation, we found that **PKN** triggers disruption of the microtubule array both in vitro and in vivo and predominantly phosphorylates tau in microtubule binding domains (MBDs). **PKN** has a catalytic domain highly homologous to protein kinase C (PKC), a kinase that phosphorylates Ser-313 (= Ser-324, the number used in this study) in MBDs. Thus, we identified the phosphorylation sites of **PKN** and PKC subtypes (PKC-alpha, -betaI, -betaII, -gamma, -delta, -epsilon, -zeta, and -lambda) in MBDs. **PKN** phosphorylates Ser-258, Ser-320, and Ser-352, although all PKC subtypes phosphorylate Ser-258, Ser-293, Ser-324, and Ser-352. There is a **PKN**-specific phosphorylation site, Ser-320, in MBDs. HIA3, a novel phosphorylation-dependent antibody recognizing phosphorylated tau at Ser-320, showed immunoreactivity in Chinese hamster ovary cells **expressing** tau and the active form of **PKN**, but not in Chinese hamster ovary cells **expressing** tau and the inactive form of **PKN**. The immunoreactivity for phosphorylated tau at Ser-320 increased in the presence of a phosphatase inhibitor, FK506 treatment, which means that calcineurin (protein phosphatase 2B) may be involved in dephosphorylating tau at Ser-320 site. We also noted that **PKN** reduces the phosphorylation recognized by the phosphorylation-dependent antibodies AT8, AT180, and AT270 in vivo. Thus **PKN** serves as a regulator of microtubules by specific phosphorylation of tau, which leads to disruption of tubulin assembly.

L10 ANSWER 24 OF 68 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2001:163310 BIOSIS
DOCUMENT NUMBER: PREV200100163310
TITLE: Regulation of gene **expression** by the small GTPase Rho through the ERK6 (p38gamma) MAP kinase pathway.
AUTHOR(S): Marinissen, Maria Julia; Chiariello, Mario; Gutkind, J. Silvio [Reprint author]
CORPORATE SOURCE: Oral and Pharyngeal Cancer Branch, National Institute of Dental and Craniofacial Research, National Institutes of Health, Bethesda, MD, 20892, USA
sg39v@nih.gov
SOURCE: Genes and Development, (March 1, 2001) Vol. 15, No. 5, pp. 535-553. print.
CODEN: GEDEEP. ISSN: 0890-9369.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 4 Apr 2001
Last Updated on STN: 15 Feb 2002

AB Small GTP-binding proteins of the Rho-family, Rho, Rac, and Cdc42, have been traditionally linked to the regulation of the cellular actin-based cytoskeleton. Rac and Cdc42 can also control the activity of JNK, thus acting in a molecular pathway transmitting extracellular signals to the nucleus. Interestingly, Rho can also regulate gene **expression**, albeit by a not fully understood mechanism. Here, we found that activated RhoA can stimulate c-jun **expression** and the activity of the c-jun promoter. As the complexity of the signaling pathways controlling the **expression** of c-jun has begun to be unraveled, this finding provided a unique opportunity to elucidate the biochemical routes whereby RhoA regulates nuclear events. We found that RhoA can initiate a linear kinase cascade leading to the activation of ERK6 (p38gamma), a recently identified member of the p38 family of MAPKs. Furthermore, we present evidence that RhoA, **PKN**, MKK3/MKK6, and ERK6 (p38gamma) are

components of a novel signal transduction pathway involved in the regulation of gene **expression** and cellular transformation.

L10 ANSWER 25 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2001:441058 HCAPLUS
DOCUMENT NUMBER: 136:211295
TITLE: Monitoring of the subtraction process in solid-phase representational difference analysis: characterization of a candidate drug
AUTHOR(S): Borang, S.; Andersson, T.; Thelin, A.; Larsson, M.; Odeberg, J.; Lundeberg, J.
CORPORATE SOURCE: Department of Biotechnology, KTH Royal Institute of Technology, Stockholm, S-100 44, Swed.
SOURCE: Gene (2001), 271(2), 183-192
CODEN: GENED6; ISSN: 0378-1119
PUBLISHER: Elsevier Science B.V.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB In this study, we have applied and evaluated a modified cDNA representational difference anal. (RDA) protocol based on magnetic bead technol. to study the mol. effects of a candidate drug (N,N'-diacetyl-L-cystine, DiNAC) in a model for atherosclerosis. Alterations in a gene **expression** profile induced by DiNAC were investigated in a **human** monocytic cell line (THP-1) differentiated into macrophage-like cells by lipopolysaccharide and further exposed to DiNAC. Three rounds of subtraction have been performed and the difference products from the second and third rounds have been characterized in detail by anal. of over 1000 gene sequences. Two protocols for anal. of the subtraction products have been evaluated, a shotgun approach and size selection of both distinct fragments and band-patterned smear. We demonstrate that in order to obtain a representative view of the most abundant gene fragments, the shotgun procedure is preferred. The obtained sequences were analyzed against the UniGene and **Expressed** Gene Anatomy Database (EGAD) databases and the results were visualized and analyzed with the ExProView software enabling rapid pair-wise comparison and identification of individual genes or functional groups of genes with altered **expression** levels. The identified differentially **expressed** gene sequences were comprised of both genes with known involvement in atherosclerosis or cholesterol biosynthesis and genes previously not implicated in these processes. The applicability of a solid-phase shotgun RDA protocol, combined with virtual chip monitoring, results in new starting points for characterization of novel candidate drugs.
REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 26 OF 68 MEDLINE on STN DUPLICATE 8
ACCESSION NUMBER: 2002018608 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11432776
TITLE: PKNbeta interacts with the SH3 domains of Graf and a novel Graf related protein, Graf2, which are GTPase activating proteins for Rho family.
AUTHOR: Shibata H; Oishi K; Yamagiwa A; Matsumoto M; Mukai H; Ono Y
CORPORATE SOURCE: Department of Biology, Faculty of Science,Graduate School of Science and Technology, Kobe University, Kobe 657-8501, Japan.
SOURCE: Journal of biochemistry, (2001 Jul) 130 (1) 23-31.
Journal code: 0376600. ISSN: 0021-924X.
PUB. COUNTRY: Japan
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-AB050785
ENTRY MONTH: 200112

ENTRY DATE: Entered STN: 20020121
Last Updated on STN: 20020121
Entered Medline: 20011205

AB PKNbeta is a novel isoform of PKNalpha, which is one of the target protein kinases for the small GTPase Rho. By yeast two-hybrid screening of a **human** embryonic kidney 293 cell cDNA library with the PKNbeta linker region containing proline-rich motifs as a bait, **clones** encoding Graf (GAP for Rho Associated with Focal adhesion kinase) and a novel Graf-related protein, termed Graf2, were isolated. The full length of Graf2 contains a putative PH domain, a RhoGAP domain, and an SH3 domain as well as Graf. Northern and Western blot analyses demonstrated that Graf2 is **expressed** in several tissues, with the highest **expression** in skeletal muscle. **Recombinant** Graf2 exhibited GTPase-activating activity toward the small GTPase RhoA and Cdc42Hs, but not toward Rac1, in vitro. The SH3 domains of Graf and Graf2 purified from Escherichia coli bound directly to PKNbeta. Graf or Graf2 was co-immunoprecipitated with PKNbeta in COS-7 cells transiently transfected with Graf or Graf2 and PKNbeta **expression** constructs. The catalytically active form of PKNbeta phosphorylated Graf and Graf2 in vitro. The interplay of PKNbeta and the GTPase-activating proteins, Graf and Graf2, may offer a novel mechanism regulating the Rho-mediated signaling.

L10 ANSWER 27 OF 68 MEDLINE on STN
ACCESSION NUMBER: 2001098534 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11006271
TITLE: Mechanism of phosphorylation of protein kinase B/Akt by a constitutively active 3-phosphoinositide-dependent protein kinase-1.
AUTHOR: Wick M J; Dong L Q; Riojas R A; Ramos F J; Liu F
CORPORATE SOURCE: Departments of Pharmacology and Biochemistry, The University of Texas Health Science Center, San Antonio, Texas 78229, USA.
CONTRACT NUMBER: DK56166 (NIDDK)
SOURCE: Journal of biological chemistry, (2000 Dec 22) 275 (51) 40400-6.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200102
ENTRY DATE: Entered STN: 20010322
Last Updated on STN: 20020420
Entered Medline: 20010201

AB Phosphorylation of Thr(308) in the activation loop and Ser(473) at the carboxyl terminus is essential for protein kinase B (PKB/Akt) activation. However, the biochemical mechanism of the phosphorylation remains to be characterized. Here we show that **expression** of a constitutively active mutant of mouse 3-phosphoinositide-dependent protein kinase-1 (PDK1(A280V)) in Chinese hamster ovary cells overexpressing the insulin receptor was sufficient to induce PKB phosphorylation at Thr(308) to approximately the same extent as insulin stimulation. Phosphorylation of PKB by PDK1(A280V) was not affected by treatment of cells with inhibitors of phosphatidylinositol 3-kinase or by deletion of the pleckstrin homology (PH) domain of PKB. C(2)-ceramide, a cell-permeable, indirect inhibitor of PKB phosphorylation, did not inhibit PDK1(A280V)-catalyzed PKB phosphorylation in cells and had no effect on PDK1 activity in vitro. On the other hand, co-**expression** of full-length protein kinase C-related kinase-1 (PRK1/**PKN**) or 2 (PRK2) inhibited PDK1(A280V)-mediated PKB phosphorylation. Replacing alanine at position 280 with valine or deletion of the PH domain enhanced PDK1 autophosphorylation in vitro. However, deletion of the PH domain of PDK1(A280V) significantly reduced PDK1(A280V)-mediated phosphorylation of

PKB in cells. In resting cells, PDK1(A280V) localized in the cytosol and at the plasma membrane. However, PDK1(A280V) lacking the PH domain localized predominantly in the cytosol. Taken together, our findings suggest that the wild-type PDK1 may not be constitutively active in cells. In addition, activation of PDK1 is sufficient to phosphorylate PKB at Thr(308) in the cytosol. Furthermore, the PH domain of PDK1 may play both positive and negative roles in regulating the in vivo function of the enzyme. Finally, unlike the carboxyl-terminal fragment of PRK2, which has been shown to bind PDK1 and allow the enzyme to phosphorylate PKB at both Thr(308) and Ser(473), full-length PRK2 and its related kinase PRK1/ **PKN** may both play negative roles in PKB-mediated downstream biological events.

L10 ANSWER 28 OF 68 MEDLINE on STN
ACCESSION NUMBER: 2001048412 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10945988
TITLE: Association of immature hypophosphorylated protein kinase cepsilon with an anchoring protein CG-NAP.
AUTHOR: Takahashi M; Mukai H; Oishi K; Isagawa T; Ono Y
CORPORATE SOURCE: Biosignal Research Center, Kobe University, Kobe 657-8501, Japan.
SOURCE: Journal of biological chemistry, (2000 Nov 3) 275 (44) 34592-6.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200012
ENTRY DATE: Entered STN: 20010322
Last Updated on STN: 20010322
Entered Medline: 20001214

AB Protein kinase C (PKC) family requires phosphorylation of itself to become competent for responding to second messengers. Much attention has been focused on elucidating the role of phosphorylation in PKC activity; however, it remains unknown where this modification takes place in the cells. This study examines whether anchoring protein is involved in the regulation of PKC phosphorylation. A certain population of PKC epsilon in rat brain extracts as well as that **expressed** in COS7 cells was associated with an endogenous anchoring protein CG-NAP (centrosome and Golgi localized **PKN**- associated protein). Pulse chase experiments revealed that the associated PKC epsilon was an immature species at the hypophosphorylated state. In vitro binding studies confirmed that non- or hypophosphorylated PKC epsilon directly bound to CG-NAP via its catalytic domain, whereas sufficiently phosphorylated PKC epsilon did not. PKC epsilon mutant at a potential phosphorylation site of Thr-566 or Ser-729 to Ala, possessing almost no catalytic activity, was associated and co-localized with CG-NAP at Golgi/centrosome area. On the other hand, wild type and a phosphorylation-mimicking mutant at Thr-566 were mainly distributed in cytosol and represented second messenger-dependent catalytic activation. These results suggest that CG-NAP anchors hypophosphorylated PKCepsilon at the Golgi/centrosome area during maturation and serves as a scaffold for the phosphorylation reaction.

L10 ANSWER 29 OF 68 MEDLINE on STN DUPLICATE 9
ACCESSION NUMBER: 2000270179 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10809724
TITLE: **PKN** binds and phosphorylates **human** papillomavirus E6 oncoprotein.
AUTHOR: Gao Q; Kumar A; Srinivasan S; Singh L; Mukai H; Ono Y; Wazer D E; Band V
CORPORATE SOURCE: Department of Radiation Oncology, New England Medical Center, Boston, Massachusetts 02111, USA.

CONTRACT NUMBER: CA64823 (NCI)
CA70195 (NCI)
SOURCE: Journal of biological chemistry, (2000 May 19) 275 (20) 14824-30.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200006
ENTRY DATE: Entered STN: 20000629
Last Updated on STN: 20000629
Entered Medline: 20000621

AB The high risk **human** papillomaviruses (HPVs) are associated with carcinomas of cervix and other genital tumors. Previous studies have identified two viral oncoproteins E6 and E7, which are **expressed** in the majority of HPV-associated carcinomas. The ability of high risk HPV E6 protein to immortalize **human** mammary epithelial cells has provided a single gene model to study the mechanisms of E6-induced oncogenic transformation. In recent years, it has become clear that in addition to E6-induced degradation of p53 tumor suppressor protein, other targets of E6 are required for mammary epithelial cells immortalization. Using the yeast two-hybrid system, we have identified a novel interaction of HPV16 E6 with protein kinase **PKN**, a fatty acid- and Rho small G protein-activated serine/threonine kinase with a catalytic domain highly homologous to protein kinase C. We demonstrate direct binding of high risk HPV E6 proteins to **PKN** in wheat-germ lysate in vitro and in 293T cells in vivo. Importantly, E6 proteins of high risk HPVs but not low risk HPVs were able to bind **PKN**. Furthermore, all the immortalization-competent and many immortalization-non-competent E6 mutants bind **PKN**. These data suggest that binding to **PKN** may be required but not sufficient for immortalizing normal mammary epithelial cells. Finally, we show that **PKN** phosphorylates E6, demonstrating for the first time that HPV E6 is a phosphoprotein. Our finding suggests a novel link between HPV E6 mediated oncogenesis and regulation of a well known phosphorylation cascade.

L10 ANSWER 30 OF 68 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
ACCESSION NUMBER: 2000:368113 SCISEARCH
THE GENUINE ARTICLE: 312MW
TITLE: Modulation of HIV-1 replication by a novel RhoA effector activity
AUTHOR: Wang L P; Zhang H C; Solski P A; Hart M J; Der C J; Su L S (Reprint)
CORPORATE SOURCE: UNIV N CAROLINA, LINEBERGER COMPREHENS CANC CTR 22 048, SCH MED, DEPT MICROBIOL & IMMUNOL, CB 7295, CHAPEL HILL, NC 27599 (Reprint); UNIV N CAROLINA, LINEBERGER COMPREHENS CANC CTR 22 048, SCH MED, DEPT MICROBIOL & IMMUNOL, CHAPEL HILL, NC 27599; UNIV N CAROLINA, LINEBERGER COMPREHENS CANC CTR, SCH MED, DEPT PHARMACOL, CHAPEL HILL, NC 27599; UNIV N CAROLINA, SCH PUBL HLTH, DEPT EPIDEMIOL, CHAPEL HILL, NC 27599; ONYX PHARMACEUT, RICHMOND, CA 94806
COUNTRY OF AUTHOR: USA
SOURCE: JOURNAL OF IMMUNOLOGY, (15 MAY 2000) Vol. 164, No. 10, pp. 5369-5374.
Publisher: AMER ASSOC IMMUNOLOGISTS, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814.
ISSN: 0022-1767.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 52

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB The RhoA GTPase is involved in regulating actin cytoskeletal

organization, gene **expression**, cell proliferation, and survival, We report here that p115-RhoGEF, a specific guanine nucleotide exchange factor (GEF) and activator of RhoA, modulates HIV-1 replication, Ectopic **expression**;of p115-RhoGEF or G alpha 13, which activates p115-RhoGEF activity, leads to inhibition of HIV-1 replication, RhoA activation is required and the inhibition affects HIV-1 gene **expression**. The RhoA effector activity in inhibiting HIV-1 replication is genetically separable from its activities in transformation of NIH3T3 cells, activation of serum response factor, and actin stress fiber formation. These findings reveal that the RhoA signal transduction pathway regulates HIV-1 replication and suggest that RhoA inhibits HIV-1 replication via a novel effector activity.

L10 ANSWER 31 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2000:468478 HCAPLUS
DOCUMENT NUMBER: 133:172515
TITLE: The Rho effector, **PKN**, regulates ANF gene transcription in cardiomyocytes through a serum response element
AUTHOR(S): Morissette, Michael R.; Sah, Valerie P.; Glembotski, Christopher C.; Brown, Joan Heller
CORPORATE SOURCE: Department of Pharmacology and Graduate Program in Biomedical Sciences, University of California, San Diego, La Jolla, CA, 92093, USA
SOURCE: American Journal of Physiology (2000), 278(6, Pt. 2), H1769-H1774
CODEN: AJPHAP; ISSN: 0002-9513
PUBLISHER: American Physiological Society
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The low-mol.-weight GTP-binding protein RhoA mediates hypertrophic growth and atrial natriuretic factor (ANF) gene **expression** in neonatal rat ventricular myocytes. Neither the effector nor the promoter elements through which Rho exerts its regulatory effects on ANF gene **expression** have been elucidated. When constitutively activated forms of Rho kinase and two protein kinase C-related kinases, **PKN** (PRK1) and PRK2, were compared, only **PKN** generated a robust stimulation of a luciferase reporter gene driven by a 638-bp fragment on the ANF promoter. This ANF promoter fragment contains a proximal serum response element (SRE) and an Sp-1-like element required for the transcriptional response to phenylephrine (PE). This response was inhibited by dominant neg. Rho. The ability of dominant neg. Rho to inhibit the response to PE and the ability of **PKN** to stimulate ANF reporter gene **expression** were both lost when the SRE was mutated. Mutation of the Sp-1-like element also attenuated the response to **PKN**. A minimal promoter driven by ANF SRE sequences was sufficient to confer Rho- and **PKN**-mediated gene **expression**. Interestingly, **PKN** preferentially stimulated the ANF vs. the c-fos SRE reporter gene. Thus **PKN** and Rho are able to regulate transcriptional activation of the ANF SRE by a common element that could implicate **PKN** as a downstream effector of Rho in transcriptional responses associated with hypertrophy.
REFERENCE COUNT: 46 THERE ARE 46 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 32 OF 68 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
ACCESSION NUMBER: 2000:848977 SCISEARCH
THE GENUINE ARTICLE: 371GG
TITLE: **Cloning** and characterization of AWP1, a novel protein that associates with serine/threonine kinase PRK1 in vivo
AUTHOR: Duan W; Sun B G; Li T W; Tan B J; Lee M K; Teo T S (Reprint)
CORPORATE SOURCE: NATL UNIV SINGAPORE, FAC MED, DEPT BIOCHEM, 10 KENT RIDGE

CRESCENT, SINGAPORE 119260, SINGAPORE (Reprint); NATL UNIV SINGAPORE, FAC MED, DEPT BIOCHEM, SINGAPORE 119260, SINGAPORE
COUNTRY OF AUTHOR: SINGAPORE
SOURCE: GENE, (3 OCT 2000) Vol. 256, No. 1-2, pp. 113-121. Publisher: ELSEVIER SCIENCE BV, PO BOX 211, 1000 AE AMSTERDAM, NETHERLANDS. ISSN: 0378-1119.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 31
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB We describe the **cloning** and **expression** of cDNAs encoding a novel **human** protein of 208 amino acid residues with a predicted molecular mass of 22.6 kDa and its mouse homologue. We name this protein as AWP1 (associated with PRK1). AWP1 is a ubiquitously **expressed** protein, and the Awp1 gene is switched on during early **human** and mouse development. When **expressed** in COS-I cells, the Myc-tagged AWP1 has an apparent molecular mass higher than that deduced from its amino acid sequence. AWP1 possesses a conserved zf-A20 zinc finger domain at its N-terminal and a zf-AN1 zinc finger domain at its C-terminal. Co-immunoprecipitation experiments revealed that mouse AWP1 specifically interacts with a rat serine/threonine protein kinase PRK1 in vivo. Hence, AWP1 may play a regulatory role in mammalian signal transduction pathways. (C) 2000 Published by Elsevier Science B.V. All rights reserved.

L10 ANSWER 33 OF 68 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2001:101558 BIOSIS
DOCUMENT NUMBER: PREV200100101558
TITLE: Inhibition of myosin phosphatase through CPI-17 phosphorylated by Rho-kinase and **protein kinase N**.
AUTHOR(S): Koyama, Mutsumi [Reprint author]; Ito, Masaaki [Reprint author]; Feng, Jianhua [Reprint author]; Seko, Tetsuya [Reprint author]; Yamawaki, Koji [Reprint author]; Isaka, Naoki [Reprint author]; Kaibuchi, Kozo; Hartshorne, David J.; Nakano, Takeshi
CORPORATE SOURCE: MIE Univ Sch of Medicine, Tsu, Japan
SOURCE: Circulation, (October 31, 2000) Vol. 102, No. 18 Supplement, pp. II.320. print. Meeting Info.: Abstracts from American Heart Association Scientific Sessions 2000. New Orleans, Louisiana, USA. November 12-15, 2000. American Heart Association. CODEN: CIRCAZ. ISSN: 0009-7322.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 28 Feb 2001
Last Updated on STN: 15 Feb 2002

L10 ANSWER 34 OF 68 MEDLINE on STN DUPLICATE 10
ACCESSION NUMBER: 1999287934 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10358086
TITLE: Characterization of a novel giant scaffolding protein, CG-NAP, that anchors multiple signaling enzymes to centrosome and the golgi apparatus.
AUTHOR: Takahashi M; Shibata H; Shimakawa M; Miyamoto M; Mukai H; Ono Y
CORPORATE SOURCE: Department of Biology, Faculty of Science, Kobe University, Kobe 657-8501, Japan.
SOURCE: Journal of biological chemistry, (1999 Jun 11) 274 (24) 17267-74.

Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-AB019691
ENTRY MONTH: 199907
ENTRY DATE: Entered STN: 19990715
Last Updated on STN: 19990715
Entered Medline: 19990706

AB A novel 450-kDa coiled-coil protein, CG-NAP (centrosome and Golgi localized **PKN**-associated protein), was identified as a protein that interacted with the regulatory region of the protein kinase **PKN**, having a catalytic domain homologous to that of protein kinase C. CG-NAP contains two sets of putative RII (regulatory subunit of protein kinase A)-binding motif. Indeed, CG-NAP tightly bound to RIIalpha in HeLa cells. Furthermore, CG-NAP was coimmunoprecipitated with the catalytic subunit of protein phosphatase 2A (PP2A), when one of the B subunit of PP2A (PR130) was exogenously **expressed** in COS7 cells. CG-NAP also interacted with the catalytic subunit of protein phosphatase 1 in HeLa cells. Immunofluorescence analysis of HeLa cells revealed that CG-NAP was localized to centrosome throughout the cell cycle, the midbody at telophase, and the Golgi apparatus at interphase, where a certain population of **PKN** and RIIalpha were found to be accumulated. These data indicate that CG-NAP serves as a novel scaffolding protein that assembles several protein kinases and phosphatases on centrosome and the Golgi apparatus, where physiological events, such as cell cycle progression and intracellular membrane traffic, may be regulated by phosphorylation state of specific protein substrates.

L10 ANSWER 35 OF 68 MEDLINE on STN DUPLICATE 11
ACCESSION NUMBER: 1999373159 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10441506
TITLE: Identification and characterization of PKNbeta, a novel isoform of protein kinase **PKN**: **expression** and arachidonic acid dependency are different from those of PKNalpha.
AUTHOR: Oishi K; Mukai H; Shibata H; Takahashi M; Ona Y
CORPORATE SOURCE: Graduate School of Science and Technology, Faculty of Science, Kobe, 657-8501, Japan.
SOURCE: Biochemical and biophysical research communications, (1999 Aug 11) 261 (3) 808-14.
Journal code: 0372516. ISSN: 0006-291X.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-AB019692
ENTRY MONTH: 199909
ENTRY DATE: Entered STN: 19990925
Last Updated on STN: 19990925
Entered Medline: 19990909

AB The cDNA **clone** encoding a novel isoform of protein kinase **PKN**, termed PKNbeta, was isolated from a HeLa cDNA library. PKNbeta had high sequence homology with PKNalpha, originally isolated **PKN**, especially in the repeats of charged amino acid-rich region with leucine-zipper like sequences (CZ region/HR1), in the carboxyl-terminal catalytic domain, and in approximately 130 amino acid stretch (D region/HR2), located between CZ region/HR1 and the catalytic domain. However, the amino acid sequence of PKNbeta differed from that of PKNalpha in the region immediately amino-terminal to the catalytic domain, which contained two distinct proline-rich sequences consistent with the class II consensus sequence, PXXPXR, for binding to SH3 domain. Distribution of PKNbeta differed from that of PKNalpha in the following

two respects: (1) Northern blotting indicated that PKNbeta mRNA could not be detected in **human** adult tissues, but was **expressed** abundantly in **human** cancer cell lines; (2) immunochemical analysis indicated that PKNbeta localized in nucleus and perinuclear Golgi apparatus, and was almost absent in cytoplasmic region in NIH3T3 cells. **Recombinant** PKNbeta **expressed** in COS7 cells displayed autophosphorylation and peptide kinase activity, but was found to be significantly less responsive to arachidonic acid than PKNalpha. The identification of this novel isoform underscores the diversity of **PKN** signaling pathway.

L10 ANSWER 36 OF 68 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2000:49012 BIOSIS
DOCUMENT NUMBER: PREV200000049012
TITLE: The structural basis of Rho effector recognition revealed by the crystal structure of **human** RhoA complexed with the effector domain of **PKN**/PRK1.
AUTHOR(S): Maesaki, Ryoko; Ihara, Kentaro; Shimizu, Toshiyuki; Kuroda, Shinya; Kaibuchi, Kozo; Hakoshima, Toshio [Reprint author]
CORPORATE SOURCE: Division of Structural Biology, Nara Institute of Science and Technology, 8916-5 Takayama, Ikoma, Nara, Japan
SOURCE: Molecular Cell, (Nov., 1999) Vol. 4, No. 5, pp. 793-803. print.
ISSN: 1097-2765.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 3 Feb 2000
Last Updated on STN: 31 Dec 2001
AB The small G protein Rho has emerged as a key regulator of cellular events involving cytoskeletal reorganization. Here we report the 2.2 ANG crystal structure of RhoA bound to an effector domain of protein kinase **PKN**/PRK1. The structure reveals the antiparallel coiled-coil finger (ACC finger) fold of the effector domain that binds to the Rho specificity-determining regions containing switch I, beta strands B2 and B3, and the C-terminal alpha helix A5, predominantly by specific hydrogen bonds. The ACC finger fold is distinct from those for other small G proteins and provides evidence for the diverse ways of effector recognition. Sequence analysis based on the structure suggests that the ACC finger fold is widespread in Rho effector proteins.

L10 ANSWER 37 OF 68 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1999:527641 BIOSIS
DOCUMENT NUMBER: PREV199900527641
TITLE: "Hit and run" targeting of the prostaglandin EP1 receptor locus introduces a point mutation which disrupts its **expression** and function.
AUTHOR(S): Qi, Zhonghua [Reprint author]; Zhang, Yahua [Reprint author]; Guan, Youfei [Reprint author]; Brandon, Suzanne [Reprint author]; Magnuson, Mark [Reprint author]; Breyer, Richard [Reprint author]; Breyer, Matthew [Reprint author]
CORPORATE SOURCE: Div. Nephrol, Vanderbilt Univ. Sch. of Medicine, Nashville, TN, USA
SOURCE: Journal of the American Society of Nephrology, (Sept., 1999) Vol. 10, No. PROGRAM AND ABSTR. ISSUE, pp. 470A. print.
Meeting Info.: 32nd Annual Meeting of the American Society of Nephrology. Miami Beach, Florida, USA. November 1-8, 1999. American Society of Nephrology.
CODEN: JASNEU. ISSN: 1046-6673.
DOCUMENT TYPE: Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 10 Dec 1999

Last Updated on STN: 10 Dec 1999

L10 ANSWER 38 OF 68 MEDLINE on STN DUPLICATE 12
ACCESSION NUMBER: 1999318776 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10388627
TITLE: Biochemical and crystallographic characterization of a Rho effector domain of the protein serine/threonine kinase N in a complex with RhoA.
AUTHOR: Maesaki R; Shimizu T; Ihara K; Kuroda S; Kaibuchi K; Hakoshima T
CORPORATE SOURCE: Division of Structural Biology, Division of Signal Transduction, Nara Institute of Science and Technology, 8916-5 Takayama, Ikoma, Nara, 630-0101, Japan.
SOURCE: Journal of structural biology, (1999 Jun 15) 126 (2) 166-70.
Journal code: 9011206. ISSN: 1047-8477.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199908
ENTRY DATE: Entered STN: 19990910
Last Updated on STN: 20000303
Entered Medline: 19990824
AB The effector domain of **human** protein serine/threonine kinase N (**PKN**), an effector protein for the small GTP-binding protein Rho, was **expressed** and purified for protein characterization and crystallization in a complex form with **human** RhoA. In solution, RhoA binds to the **PKN** effector domain with 1:2 stoichiometry in a GTP-dependent manner. The obtained complex crystals diffract to 2.2 A resolution.

L10 ANSWER 39 OF 68 MEDLINE on STN DUPLICATE 13
ACCESSION NUMBER: 2000109325 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10640683
TITLE: Interaction of **PKN** with a neuron-specific basic helix-loop-helix transcription factor, NDRF/NeuroD2.
AUTHOR: Shibata H; Oda H; Mukai H; Oishi K; Misaki K; Ohkubo H; Ono Y
CORPORATE SOURCE: Department of Biology, Faculty of Science, Kobe University, Kobe, Japan.
SOURCE: Brain research. Molecular brain research, (1999 Dec 10) 74 (1-2) 126-34.
Journal code: 8908640. ISSN: 0169-328X.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200002
ENTRY DATE: Entered STN: 20000229
Last Updated on STN: 20000229
Entered Medline: 20000215
AB By the yeast two-hybrid screening of a **human** brain cDNA library with the amino-terminal regulatory region of **PKN** as a bait, a **clone** encoding a neuron-specific basic Helix-Loop-Helix (bHLH) transcription factor, NDRF/NeuroD2 was isolated. NDRF/NeuroD2 was co-precipitated with **PKN** from the lysate of COS-7 cells transfected with both **expression** constructs for NDRF/NeuroD2 and **PKN**. In vitro binding studies using the deletion mutants of NDRF/NeuroD2 synthesized in a rabbit reticulocyte lysate indicated that the internal region containing the bHLH domain of NDRF/NeuroD2 was necessary and sufficient for the interaction with **PKN**. In addition, **recombinant** NDRF/NeuroD2 purified from Escherichia

coli could bind **PKN**, suggesting the direct interaction between NDRF/NeuroD2 and **PKN**. Transient transfection assays using P19 cells revealed that **expression** of NDRF/NeuroD2 increased the transactivation of the rat insulin promoter element 3 (RIPE3) enhancer up to approximately 12-fold and that co-**expression** of catalytically active form of **PKN**, but not kinase-deficient derivative, resulted in a further threefold increase of NDRF/NeuroD2-mediated transcription. These findings suggest that **PKN** may contribute to transcriptional responses through the post-translational modification of the NDRF/NeuroD2-dependent transcriptional machinery.

L10 ANSWER 40 OF 68 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN DUPLICATE 14

ACCESSION NUMBER: 1998266576 EMBASE
TITLE: Different regions of Rho determine Rho-selective binding of different classes of Rho target molecules.
AUTHOR: Fujisawa K.; Madaule P.; Ishizaki T.; Watanabe G.; Bito H.; Saito Y.; Hall A.; Narumiya S.
CORPORATE SOURCE: S. Narumiya, Dept. of Pharmacology, Kyoto University Faculty of Medicine, Yoshida, Sakyo-ku, Kyoto 606-8315, Japan. snaru@mfour.med.kyoto-u.ac.jp
SOURCE: Journal of Biological Chemistry, (24 Jul 1998) 273/30 (18943-18949).
Refs: 45
ISSN: 0021-9258 CODEN: JBCHA3
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Based on their Rho binding motifs several Rho target molecules can be classified into three groups; class I includes the protein kinase **PKN**, rhophilin, and rhotekin, class II includes the protein kinases, Rho- associated coiled-coil containing protein kinases, ROCK-I and ROCK-II, and class HI includes citron. Taking advantage of the selectivity in recognition by these targets between Rho and Rac, we examined the regions in Rho required for selective binding of each class of Rho target molecules. Yeast two- hybrid assays were performed using Rho/Rac chimeras and either rhophilin, ROCK-I, or citron. This study showed the existence of at least two distinct regions in Rho (amino acids 23-40 and 75-92) that are critical for the selective binding of these targets. The former was required for binding to citron, whereas the latter was necessary for binding to rhophilin. On the other hand, either region showed affinity to ROCK-I. This was further confirmed by ligand overlay assay using both **recombinant** ROCK-I and ROCK-II proteins. Consistently, Rho/Rac chimeras containing either region can induce stress fibers in transfected HeLa cells, and this induction is suppressed by treatment with Y-27632, a specific inhibitor of ROCK kinases. These results suggest that the selective binding of different classes of Rho targets to Rho is determined by interaction between distinct Rho-binding motifs of the targets and different regions of Rho.

L10 ANSWER 41 OF 68 MEDLINE on STN DUPLICATE 15

ACCESSION NUMBER: 1998426194 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9751706
TITLE: Proteolytic activation of **PKN** by caspase-3 or related protease during apoptosis.
AUTHOR: Takahashi M; Mukai H; Toshimori M; Miyamoto M; Ono Y
CORPORATE SOURCE: Department of Biology, Faculty of Science, Kobe University, Kobe 657-8501, Japan.
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (1998 Sep 29) 95 (20) 11566-71.
Journal code: 7505876. ISSN: 0027-8424.
PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199810
ENTRY DATE: Entered STN: 19981029
Last Updated on STN: 20000303
Entered Medline: 19981022

AB **PKN**, a fatty acid- and Rho-activated serine/threonine kinase having a catalytic domain highly homologous to protein kinase C (PKC), was cleaved at specific sites in apoptotic Jurkat and U937 cells on Fas ligation and treatment with staurosporin or etoposide, respectively. The cleavage of **PKN** occurred with a time course similar to that of PKCdelta, a known caspase substrate. This proteolysis was inhibited by a caspase inhibitor, acetyl-Asp-Glu-Val-Asp-aldehyde. The cleavage fragments were generated in vitro from **PKN** by treatment with **recombinant** caspase-3. Site-directed mutagenesis of specific aspartate residues prevented the appearance of these fragments. These results indicate that **PKN** is cleaved by caspase-3 or related protease during apoptosis. The major proteolysis took place between the amino-terminal regulatory domain and the carboxyl-terminal catalytic domain, and it generated a constitutively active kinase fragment. The cleavage of **PKN** may contribute to signal transduction, eventually leading to apoptosis.

L10 ANSWER 42 OF 68 MEDLINE on STN
ACCESSION NUMBER: 1998112814 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9446575
TITLE: Multiple interactions of PRK1 with RhoA. Functional assignment of the Hr1 repeat motif.
AUTHOR: Flynn P; Mellor H; Palmer R; Panayotou G; Parker P J
CORPORATE SOURCE: Protein Phosphorylation Laboratory, Imperial Cancer Research Fund, 44 Lincoln's Inn Fields, London WC2A 3PX, United Kingdom.
SOURCE: Journal of biological chemistry, (1998 Jan 30) 273 (5) 2698-705.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199802
ENTRY DATE: Entered STN: 19980306
Last Updated on STN: 20000303
Entered Medline: 19980223

AB PRK1 (**PKN**) is a serine/threonine kinase that has been shown to be activated by RhoA (Amano, M., Mukai, H., Ono, Y., Chihara, K., Matsui, T., Hamajima, Y., Okawa, K., Iwamatsu, A., and Kaibuchi, K. (1996) Science 271, 648-650). Detailed analysis of the PRK1 region involved in RhoA binding has revealed that two homologous sequences within the HR1 domain (HR1a and HR1b) both bind to RhoA; the third repeat within this domain, HR1cPRK1, does not bind RhoA. The related HR1 motif is also found to confer RhoA binding activity to the only other fully **cloned** member of this kinase family, PRK2. Furthermore, the predictive value of this motif is established for an HR1a sequence derived from a Caenorhabditis elegans open reading frame encoding a protein kinase of unknown function. Interestingly, the HR1aPRK1 and HR1bPRK1 subdomains are shown to display a distinctive nucleotide dependence for RhoA binding. HRIaPRK1 is entirely GTP-dependent, while HR1bPRK1 binds both GTP- and GDP-bound forms of RhoA. This distinction indicates that there are two sites of contact between RhoA and PRK1, one contact through a region that is conformationally dependent upon the nucleotide-bound state of RhoA and one that is not. Analysis of binding to Rho/Rac chimera provides evidence for a HR1aPRK1 but not HR1bPRK1 interaction in the central third of Rho. Additionally, it is observed that the V14RhoA mutant binds HR1a but does

not bind HR1b. This distinct binding behavior corroborates the conclusion that there are independent contacts on RhoA for the HR1aPRK1 and HR1bPRK1 motifs.

L10 ANSWER 43 OF 68 MEDLINE on STN
ACCESSION NUMBER: 1999106268 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9889594
TITLE: Protein kinase **PKN**.
AUTHOR: Mukai H; Ono Y
CORPORATE SOURCE: Graduate School of Science and Technology, Kobe University.
SOURCE: Seikagaku. Journal of Japanese Biochemical Society, (1998 Nov) 70 (11) 1335-9. Ref: 16
Journal code: 0413564. ISSN: 0037-1017.
PUB. COUNTRY: Japan
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: Japanese
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199903
ENTRY DATE: Entered STN: 19990402
Last Updated on STN: 20000303
Entered Medline: 19990325

L10 ANSWER 44 OF 68 MEDLINE on STN DUPLICATE 16
ACCESSION NUMBER: 1999057541 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9837746
TITLE: The role of **PKN** in the regulation of alphaB-crystallin **expression** via heat shock transcription factor 1.
AUTHOR: Kitagawa M; Mukai H; Takahashi M; Ono Y
CORPORATE SOURCE: Graduate School of Science and Technology, Faculty of Science, Kobe, 657-8501, Japan.
SOURCE: Biochemical and biophysical research communications, (1998 Nov 27) 252 (3) 561-5.
Journal code: 0372516. ISSN: 0006-291X.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199901
ENTRY DATE: Entered STN: 19990128
Last Updated on STN: 19990128
Entered Medline: 19990114

AB We previously reported that **PKN**, a fatty acid-activated serine/threonine protein kinase, translocates from the cytosol to the nucleus by stresses such as heat shock, sodium arsenite, and serum starvation. To clarify the role of **PKN** under heat stress, we examined whether **PKN** regulates the **expression** of heat shock proteins. Co-**expression** of heat shock transcription factor 1 (HSF1) and the catalytically active fragment of **PKN** induced the accumulation of alphaB-crystallin but not HSP27 and HSP70 in HeLa S3 cells. The **expression** of the reporter gene for alphaB-crystallin promoter was activated by co-**expression** of HSF1 and the catalytically active fragment of **PKN**, and this activation was dependent on the protein kinase activity of **PKN**. Deletion analysis of the alphaB-crystallin promoter region revealed that both the proximal and the distal heat shock elements were necessary for the transactivation. These results raise the possibility that there is a signal transduction pathway mediating stress signals for the accumulation of alphaB-crystallin by HSF1 and **PKN**.

L10 ANSWER 45 OF 68 MEDLINE on STN DUPLICATE 17

ACCESSION NUMBER: 1998303811 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9637778
TITLE: **PKN** interacts with a paraneoplastic cerebellar degeneration-associated antigen, which is a potential transcription factor.
AUTHOR: Takanaga H; Mukai H; Shibata H; Toshimori M; Ono Y
CORPORATE SOURCE: Faculty of Science, Kobe University, Kobe, 657, Japan.
SOURCE: Experimental cell research, (1998 Jun 15) 241 (2) 363-72. Journal code: 0373226. ISSN: 0014-4827.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199807
ENTRY DATE: Entered STN: 19980723
Last Updated on STN: 19980723
Entered Medline: 19980714

AB **PKN** is a fatty acid-activated serine/threonine protein kinase, having a catalytic domain homologous to protein kinase C family. **PKN** has been recently reported to interact with a small GTP-binding protein Rho and cytoskeletal proteins such as neurofilament and alpha-actinin. To identify the new components of the **PKN** -signaling pathway, the yeast two-hybrid system was employed. Using the amino-terminal regulatory domain of **PKN** as a bait, cDNA encoding a neural antigen PCD17, which is recognized by characteristic antibodies of patients with paraneoplastic cerebellar degeneration, was isolated from a **human** brain cDNA library. The interaction between **PKN** and PCD17 was also determined by the in vitro binding analysis. PCD17 was coimmunoprecipitated with **PKN** from the lysate of COS7 cells transfected with both **expression** constructs for **PKN** and the amino-terminal region of PCD17. PCD17 was phosphorylated by **PKN**, and the extent of this phosphorylation was enhanced by addition of 40 microM arachidonic acid. The amino-terminal region of PCD17 could form a homodimer in vitro, and PCD17 fused to the Gal4 DNA binding domain showed the transcriptional transactivation of the chloramphenicol acetyltransferase reporter gene linked to 5 Gal4 binding sites and minimal promoter in rat C6 glioma cells. These results suggest the participation of PCD17 in gene **expression** and lead to a clue for elucidating the **PKN** signaling pathway from the cytosol to the nucleus.

L10 ANSWER 46 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1998:10482 HCAPLUS
DOCUMENT NUMBER: 128:125301
TITLE: Characterization and use of **protein kinase N** derivatives capable of inhibiting the binding between **protein kinase N** and activated Rho family proteins
INVENTOR(S): Kaibuchi, Kozo; Ono, Isataka; Iwamatsu, Akihiko
PATENT ASSIGNEE(S): Kirin Brewery Co., Ltd., Japan
SOURCE: Jpn. Kokai Tokkyo Koho, 68 pp.
CODEN: JKXXAF
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 09327292	A2	19971222	JP 1996-213245	19960724
US 6660837	B1	20031209	US 1996-685852	19960724

PRIORITY APPLN. INFO.: JP 1995-262552 A 19950914

JP 1995-344606 A 19951205
JP 1996-80549 A 19960308
JP 1996-114226 A 19960411

AB Disclosed are **protein kinase N** (**PKN**) derivs. capable of inhibiting the binding between **protein kinase N** and activated Rho family proteins. The derivs., that are able to bind with Rho proteins but lacking protein kinase activities, are prepared by changing the amino acid sequence (chemical modification of side chains, substitution, deletion, etc.) of **human PKN** or its fragments of, e.g., amino acid number 7.apprx.540, 7.apprx.155, 1.apprx.538, 3.apprx.135, 33.apprx.111, 74.apprx.93, 94.apprx.113, and 82.apprx.103. The **PKN** derivs. capable of inhibiting the binding between **PKN** and intermediate filaments or α-actinin, or inhibiting the transport of **PKN** from cytoplasm to nucleus are also described. Also claimed are derivs. or fragments of α-actinin or intermediate filaments that able to bind to **PKN**, methods of **recombinant** preparation of **PKN** derivs., use of the **PKN** derivs. for treating tumor metastasis in gene therapy, and methods of screening Rho protein GTPase activity-inhibitory substances, **PKN**-intermediate filaments binding-inhibitory substances, intermediate filaments polymerization-inhibitory substances, or **PKN**-α-actinin binding-inhibitory substances.

L10 ANSWER 47 OF 68 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
ACCESSION NUMBER: 97:874824 SCISEARCH
THE GENUINE ARTICLE: YG647
TITLE: Specific proteolysis of the kinase protein kinase C-related kinase 2 by caspase-3 during apoptosis - Identification by a novel, small pool **expression cloning** strategy
AUTHOR: Cryns V L; Byun Y; Rana A; Mellor H; Lustig K D; Ghanem L; Parker P J; Kirschner M W; Yuan J Y (Reprint)
CORPORATE SOURCE: HARVARD UNIV, SCH MED, DEPT CELL BIOL, LHRRB 409, 240 LONGWOOD AVE, BOSTON, MA 02115 (Reprint); HARVARD UNIV, SCH MED, DEPT CELL BIOL, BOSTON, MA 02115; MASSACHUSETTS GEN HOSP, DIABET UNIT, CHARLESTOWN, MA 02129; IMPERIAL CANC RES FUND, PROT PHOSPHORYLAT LAB, LONDON WC2A 3PX, ENGLAND
COUNTRY OF AUTHOR: USA; ENGLAND
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (21 NOV 1997) Vol. 272, No. 47, pp. 29449-29453.
Publisher: AMER SOC BIOCHEMISTRY MOLECULAR BIOLOGY INC, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814.
ISSN: 0021-9258.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 26
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB The caspase family of proteases plays a critical role in the execution of apoptosis, However, efforts to decipher the molecular mechanisms by which caspases induce cell death have been greatly hindered by the lack of systematic and broadly applicable strategies to identify their substrates. Here we describe a novel **expression cloning** strategy to rapidly isolate cDNAs encoding caspase substrates that are cleaved during apoptosis, Small cDNA pools (approximately 100 **clones** each) are transcribed/translated in vitro in the presence of [S-35]methionine; these labeled protein pools are then incubated with cytosolic extracts from control and apoptotic cells, cDNA pools encoding proteins that are specifically cleaved by the apoptotic extract and whose cleavage is prevented by the caspase inhibitor acetyl-Tyr-Val-Ala-Asp chloromethylketone are subdivided and retested until a single cDNA is isolated, Using this approach, we isolated a partial cDNA encoding protein

kinase C-related kinase 2 (PRK2), a serine-threonine kinase, and demonstrate that full-length **human** PRK2 is proteolyzed by caspase-3 at Asp(117) and Asp(700) in vitro, In addition, PRK2 is cleaved rapidly during Fas-and staurosporine-induced apoptosis in vitro by caspase-3 or a closely related caspase, Both of the major apoptotic cleavage sites of PRK2 in vivo lie within its regulatory domain, suggesting that its activity may be deregulated by proteolysis.

L10 ANSWER 48 OF 68 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
DUPLICATE 18
ACCESSION NUMBER: 1997:225059 BIOSIS
DOCUMENT NUMBER: PREV199799516775
TITLE: Isolation and characterization of a structural homologue of **human** PRK2 from rat liver. Distinguishing substrate and lipid activator specificities.
AUTHOR(S): Yu, Weiping; Liu, Junjun; Morrice, Nicholas A.; Wettenhall, Richard E. H. [Reprint author]
CORPORATE SOURCE: Russell Grimwade Sch. Biochem. Mol. Biol., Univ. Melbourne, Parkville, VIC 3052, Australia
SOURCE: Journal of Biological Chemistry, (1997) Vol. 272, No. 15, pp. 10030-10034.
CODEN: JBCHA3. ISSN: 0021-9258.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 22 May 1997
Last Updated on STN: 9 Jul 1997
AB A homologue of **human** protein C (PKC)-related kinase-2, PRK2, which had previously escaped identification in normal mammalian tissues, was isolated from rat liver as the protease-activated kinase (PAK) originally named PAK-2. The 130-kDa cytosolic enzyme was purified to homogeneity and shown by tryptic peptide and reverse transcriptase-polymerase chain reaction (RT-PCR)-amplified rat cDNA sequence analyses to be structurally related to the 116-kDa rat hepatic PAK-1/**protein kinase N** (**PKN**) and, even more closely (95% sequence identity) to the 130-kDa **human** PKC-related kinase, PRK2. Rat myeloma RNA was used as the RT-PCR template because of its relative abundance in PAK-2/PRK2 mRNA compared with liver and other rat tissues. The catalytic properties of PAK-2/PRK2 in many respects resembled those of hepatic PAK-1/**PKN**, but were distinguished by more favorable kinetics with several peptide substrates, and greater sensitivity to PKC pseudosubstrate and polybasic amino acid inhibitors. PAK-2/PRK2 was also activated by lipids, particularly cardiolipin and to a lesser extent by other acidic phospholipids and unsaturated fatty acids. Cardiolipin activation was most evident with autophosphorylation and histone H2B phosphorylation, but only marginally evident with the favored ribosomal S6-(229-239) peptide substrate for the protease-activated kinase activity. It was concluded that PAK-2 is the rat homologue of **human** PRK2, with biochemical properties distinct from although overlapping those of the PAK-1/**PKN**/PRK1 isoform.

L10 ANSWER 49 OF 68 MEDLINE on STN DUPLICATE 19
ACCESSION NUMBER: 97184114 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9030526
TITLE: Interaction of **PKN** with alpha-actinin.
AUTHOR: Mukai H; Toshimori M; Shibata H; Takanaga H; Kitagawa M; Miyahara M; Shimakawa M; Ono Y
CORPORATE SOURCE: Radioisotope Research Center, Kobe University, Kobe 657, Japan.
SOURCE: Journal of biological chemistry, (1997 Feb 21) 272 (8) 4740-6.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English

FILE SEGMENT: Priority Journals
ENTRY MONTH: 199704
ENTRY DATE: Entered STN: 19970414
Last Updated on STN: 19970414
Entered Medline: 19970403

AB **PKN** is a fatty acid- and Rho-activated serine/threonine protein kinase, having a catalytic domain homologous to protein kinase C family. To identify components of the **PKN**-signaling pathway such as substrates and regulatory proteins of **PKN**, the yeast two-hybrid strategy was employed. Using the N-terminal region of **PKN** as a bait, cDNAs encoding actin cross-linking protein alpha-actinin, which lacked the N-terminal actin-binding domain, were isolated from **human** brain cDNA library. The responsible region for interaction between **PKN** and alpha-actinin was determined by in vitro binding analysis using the various truncated mutants of these proteins. The N-terminal region of **PKN** outside the RhoA-binding domain was sufficiently shown to associate with alpha-actinin. **PKN** bound to the third spectrin-like repeats of both skeletal and non-skeletal muscle type alpha-actinin. **PKN** also bound to the region containing EF-hand-like motifs of non-skeletal muscle type alpha-actinin in a Ca2+-sensitive manner and bound to that of skeletal muscle type alpha-actinin in a Ca2+-insensitive manner. alpha-Actinin was co-immunoprecipitated with **PKN** from the lysate of COS7 cells transfected with both **expression** constructs for **PKN** and alpha-actinin lacking the actin-binding domain. In vitro translated full-length alpha-actinin containing the actin-binding site hardly bound to **PKN**, but the addition of phosphatidylinositol 4, 5-bisphosphate, which is implicated in actin reorganization, stimulated the binding activity of the full-length alpha-actinin with **PKN**. We therefore propose that **PKN** is linked to the cytoskeletal network via a direct association between **PKN** and alpha-actinin.

L10 ANSWER 50 OF 68 MEDLINE on STN
ACCESSION NUMBER: 97318826 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9175763
TITLE: Domain-specific phosphorylation of vimentin and glial fibrillary acidic protein by **PKN**.
AUTHOR: Matsuzawa K; Kosako H; Inagaki N; Shibata H; Mukai H; Ono Y; Amano M; Kaibuchi K; Matsuura Y; Azuma I; Inagaki M
CORPORATE SOURCE: Laboratory of Biochemistry, Aichi Cancer Center Research Institute, Nagoya, Japan.
SOURCE: Biochemical and biophysical research communications, (1997 May 29) 234 (3) 621-5.
Journal code: 0372516. ISSN: 0006-291X.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199706
ENTRY DATE: Entered STN: 19970716
Last Updated on STN: 19970716
Entered Medline: 19970630

AB **PKN** is a serine/threonine protein kinase with a catalytic domain homologous to the protein kinase C family and unique N-terminal leucine zipper-like sequences. Using analyses with the yeast two-hybrid system and in vitro binding assay, we found that the regulatory domain of **PKN** interacted with vimentin. We then examined whether **PKN** would phosphorylate vimentin in vitro. Vimentin proved to be an excellent substrate for **PKN**, and the phosphorylation of vimentin by **PKN** occurred in the head domain with the result of a nearly complete inhibition of its filament formation in vitro. Similar results were also obtained with another type III intermediate filament protein, glial fibrillary acidic protein (GFAP). These results raise the possibility that **PKN** may regulate filament structures of

vimentin and GFAP by domain-specific phosphorylation.

L10 ANSWER 51 OF 68 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1998:46393 BIOSIS
DOCUMENT NUMBER: PREV199800046393
TITLE: Interaction of the **human** protein kinase PKR with the mouse PKR homolog occurs via the N-terminal region of PKR and does not inactivate autophosphorylation activity of mouse PKR.
AUTHOR(S): Rende-Fournier, Rosanna; Ortega, Laura G.; George, Cyril X.; Samuel, Charles E. [Reprint author]
CORPORATE SOURCE: Interdepartmental Graduate Program Biochemistry, Mol. Biol., Univ. Calif., Santa Barbara, CA 93106, USA
SOURCE: Virology, (Nov. 24, 1997) Vol. 238, No. 2, pp. 410-423. print.
CODEN: VIRLAX. ISSN: 0042-6822.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 27 Jan 1998
Last Updated on STN: 27 Jan 1998

AB The RNA-dependent protein kinase (PKR) is implicated in the antiviral and antiproliferative actions of interferon. Mutant forms of **human** PKR display a transdominant behavior when **expressed** in transfected cells. The potential for the **human** PKR protein to physically interact with the mouse PKR homolog has therefore been examined. The yeast two-hybrid system was used to probe the association between mouse and **human** PKR proteins as measured by activation of two Gal4-responsive reporter genes, HIS3 and lacZ. **Expression** of full-length wild-type mouse PKR(1-515)WT as a Gal4 fusion protein did not exhibit the growth suppression phenotype in yeast characteristic of wild-type **human** PKR(1-551)WT. Coexpression of mouse PKR(1-515)WT as a Gal4 DNA-binding domain fusion with either the catalytic-deficient **human** PKR(1-551) K296R mutant, the RNA-binding-deficient **human** PKR(1-551)K64E/K296R double mutant, or wild-type mouse PKR(1-515)WT as full-length PKR-Gal4 activation domain fusions resulted in activation of the HIS3 and lacZ reporters. The N-terminal RNA-binding region of **human** PKR, both WT and the K64E RNA-binding-deficient mutant, also interacted with mouse PKR(1-515)WT sufficiently to activate the reporters but the **human** catalytic region did not. Mouse and **human** full-length PKR proteins **expressed** as glutathione S-transferase (GST) fusions in Escherichia coli were purified on Sepharose beads. Using GST-PKR fusion chromatography, direct physical interaction between the mouse and **human** PKR homologs was established. Intraspecies PKR interactions were more efficient than interspecies PKR interactions, and interactions between RNA-binding-sufficient PKR proteins were more efficient than those involving an RNA-binding mutant as measured by binding to GST-PKR protein Sepharose beads. The N-terminal region of **human** PKR within amino acids 1-184 was sufficient for binding mouse PKR. Purified mouse full-length PKR(1-515)WT GST fusion protein retained kinase activity on Sepharose beads, but the activity was not impaired by association with either the full-length or the N-terminal region of **human** PKR.

L10 ANSWER 52 OF 68 MEDLINE on STN
ACCESSION NUMBER: 97236294 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9125115
TITLE: Identification of a novel Drosophila protein kinase highly homologous to **protein kinase N** (**PKN**).
AUTHOR: Ueno N; Oishi I; Sugiyama S; Nishida Y; Minami Y; Yamamura H
CORPORATE SOURCE: Department of Biochemistry, Kobe University School of Medicine, Japan.
SOURCE: Biochemical and biophysical research communications, (1997

Mar 6) 232 (1) 126-9.
Journal code: 0372516. ISSN: 0006-291X.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK
ENTRY MONTH: 199704
ENTRY DATE: Entered STN: 19970506
Last Updated on STN: 19970506
Entered Medline: 19970422

AB We identified a novel Drosophila gene, Dpkn (Drosophila protein kinase related to **PKN**), encoding a putative protein serine/threonine kinase. Although the cDNA obtained was incomplete at its 5'-terminal region, the deduced amino acid sequence of its kinase domain exhibits a high degree of similarity to **protein kinase N** (**PKN**), which has a kinase domain related to protein kinase C (PKC) and leucine zipper-like sequences in the amino terminal region. **Expression** of Dpkn was observed throughout Drosophila development, although its **expression** level decreased at later stages of embryogenesis. The **expression** of Dpkn is first detected in the newly formed mesodermal cell layer and is then restricted to the developing somatic musculature, indicating a possible role of Dpkn in the development of somatic muscles in Drosophila.

L10 ANSWER 53 OF 68 MEDLINE on STN
ACCESSION NUMBER: 96199250 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8621664
TITLE: **PKN** associates and phosphorylates the head-rod domain of neurofilament protein.
AUTHOR: Mukai H; Toshimori M; Shibata H; Kitagawa M; Shimakawa M; Miyahara M; Sunakawa H; Ono Y
CORPORATE SOURCE: Department of Biology, Faculty of Science, Kobe University, Japan.
SOURCE: Journal of biological chemistry, (1996 Apr 19) 271 (16) 9816-22.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199606
ENTRY DATE: Entered STN: 19960627
Last Updated on STN: 19980206
Entered Medline: 19960618

AB **PKN** is a fatty acid-activated serine/threonine kinase that has a catalytic domain highly homologous to that of protein kinase C in the carboxyl terminus and a unique regulatory region in the amino terminus. Recently, we reported that the small GTP-binding protein Rho binds to the amino-terminal region of **PKN** and activates **PKN** in a GTP-dependent manner, and we suggested that **PKN** is located on the downstream of Rho in the signal transduction pathway (Amano, M., Mukai, H., Ono, Y., Chihara, K., Matsui, T., Hamajima, Y., Okawa, K., Iwamatsu, A., and Kaibuchi, K. (1996) Science 271, 648-650; Watanabe, G., Saito, Y., Madaule, P., Ishizaki, T., Fujisawa, K., Morii, N., Mukai, H., Ono, Y. Kakizuka, A., and Narumiya, S. (1996) Science 271, 645-648). To identify other components of the **PKN** pathway such as substrates and regulatory proteins of **PKN**, the yeast two-hybrid strategy was employed. By this screening, a **clone** encoding the neurofilament L protein, a subunit of neuron-specific intermediate filament, was isolated. The amino-terminal regulatory region of **PKN** was shown to associate with the head-rod domains of other subunits of neurofilament (neurofilament proteins M and H) as well as neurofilament L protein in yeast cells. The direct binding between

PKN and each subunit of neurofilament was confirmed by using the in vitro translated amino-terminal region of **PKN** and glutathione S-transferase fusion protein containing the head-rod domain of each subunit of neurofilament. **PKN** purified from rat testis phosphorylated each subunit of the native neurofilament purified from bovine spinal cord and the bacterially synthesized head-rod domain of each subunit of neurofilament. Polymerization of neurofilament L protein in vitro was inhibited by phosphorylation of neurofilament L protein by **PKN**. The identification and characterization of the novel interaction with **PKN** may contribute toward the elucidation of mechanisms regulating the function of neurofilament.

L10 ANSWER 54 OF 68 MEDLINE on STN DUPLICATE 20
ACCESSION NUMBER: 96183060 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8607876
TITLE: The role of the unique motifs in the amino-terminal region of **PKN** on its enzymatic activity.
AUTHOR: Kitagawa M; Shibata H; Toshimori M; Mukai H; Ono Y
CORPORATE SOURCE: Department of Biology, Faculty of Science, Kobe University, Japan.
SOURCE: Biochemical and biophysical research communications, (1996 Mar 27) 220 (3) 963-8.
Journal code: 0372516. ISSN: 0006-291X.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199605
ENTRY DATE: Entered STN: 19960531
Last Updated on STN: 19970203
Entered Medline: 19960520
AB The yeast two-hybrid system and in vitro binding assay were carried out to characterize the interaction between the amino-terminal and carboxyl-terminal region of **PKN**. It was revealed that the amino-terminal region containing the regulatory domain associated with the carboxyl-terminal catalytic region. A synthetic peptide, corresponding to the amino acid residues of **PKN** from 39 to 53, with substitution of isoleucine46 with serine was shown to become a potent substrate for **PKN**, and its wild type synthetic peptide inhibited the phosphorylation by **PKN**. These results suggest that the amino-terminal region of **PKN** contains the pseudosubstrate sequence and acts as an autoinhibitory region.

L10 ANSWER 55 OF 68 MEDLINE on STN DUPLICATE 21
ACCESSION NUMBER: 96165390 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8571126
TITLE: **Protein kinase N** (**PKN**) and **PKN**-related protein rhophilin as targets of small GTPase Rho.
AUTHOR: Watanabe G; Saito Y; Madaule P; Ishizaki T; Fujisawa K; Morii N; Mukai H; Ono Y; Kakizuka A; Narumiya S
CORPORATE SOURCE: Department of Pharmacology, Kyoto University Faculty of Medicine, Japan.
SOURCE: Science, (1996 Feb 2) 271 (5249) 645-8.
Journal code: 0404511. ISSN: 0036-8075.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-U43194
ENTRY MONTH: 199603
ENTRY DATE: Entered STN: 19960315
Last Updated on STN: 20000303
Entered Medline: 19960305

AB The Rho guanosine 5'-triphosphatase (GTPase) cycles between the active guanosine triphosphate (GTP)-bound form and the inactive guanosine diphosphate-bound form and regulates cell adhesion and cytokinesis, but how it exerts these actions is unknown. The yeast two-hybrid system was used to **clone** a complementary DNA for a protein (designated Rhophilin) that specifically bound to GTP-Rho. The Rho-binding domain of this protein has 40 percent identity with a putative regulatory domain of a protein kinase, **PKN**. **PKN** itself bound to GTP-Rho and was activated by this binding both in vitro and in vivo. This study indicates that a serine-threonine protein kinase is a Rho effector and presents an amino acid sequence motif for binding to GTP-Rho that may be shared by a family of Rho target proteins.

L10 ANSWER 56 OF 68 MEDLINE on STN
ACCESSION NUMBER: 96359842 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8719789
TITLE: The pharmacology of GR203040, a novel, potent and selective non-peptide tachykinin NK1 receptor antagonist.
AUTHOR: Beattie D T; Beresford I J; Connor H E; Marshall F H; Hawcock A B; Hagan R M; Bowers J; Birch P J; Ward P
CORPORATE SOURCE: Pharmacology II Department, Glaxo Wellcome Medicines Research Centre, Stevenage, Herts.
SOURCE: British journal of pharmacology, (1995 Dec) 116 (8) 3149-57.
Journal code: 7502536. ISSN: 0007-1188.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199610
ENTRY DATE: Entered STN: 19961022
Last Updated on STN: 19980206
Entered Medline: 19961010

AB 1. The in vitro and in vivo pharmacology of GR203040 ((2S, 3S)-2-methoxy-5-tetrazol-1-yl-benzyl-(2-phenyl-piperidin-3-y l)-amine), a novel, highly potent and selective non-peptide tachykinin NK1 receptor antagonist, was investigated in the present study. 2. GR203040 potently inhibited [3H]-substance P binding to **human** NK1 receptors **expressed** in Chinese hamster ovary (CHO) and U373 MG astrocytoma cells, and NK1 receptors in ferret and gerbil cortex (pKi values of 10.3, 10.5, 10.1 and 10.1 respectively). GR203040 had lower affinity at rat NK1 receptors (pKi = 8.6) and little affinity for **human** NK2 receptors (pKi < 5.0) in CHO cells and NK3 receptors in guinea-pig cortex (pKi < 6.0). With the exception of the histamine H1 receptor (pIC50 = 7.5). GR203040 had little affinity (pIC50 < 6.0) at all non-NK1 receptors and ion channels examined. Furthermore, GR203040 produced only weak inhibition of Na+ currents in SH-SY5Y neuroblastoma and superior cervical ganglion cells (pIC50 values < 4.0). GR203040 produced only weak antagonism of Ca(2+)-evoked contractions of rat isolated portal vein (**pKn** = 4.1). The enantiomer of GR203040, GR205608 (2R, 3R)-2-methoxy-5-tetrazol-1-yl-benzyl-(2-phenyl-piperidin-3-y l)-amine), had 10,000 fold lower affinity at the **human** NK1 receptor **expressed** in CHO cells (pKi = 6.3). 3. In gerbil ex vivo binding experiments, GR203040 produced a dose-dependent inhibition of the binding of [3H]-substance P to cerebral cortical membranes (ED50 = 15 micrograms kg-1 s.c. and 0.42 mg kg-1 p.o.). At 10 micrograms kg-1 s.c., the inhibition of [3H]-substance P binding was maintained for > 6 h. In the rat, GR203040 was less potent (ED50 = 15.4 mg kg-1 s.c.) probably reflecting, at least in part, its lower affinity at the rat NK1 receptor. 4. In guinea-pig isolated ileum and dog isolated middle cerebral and basilar arteries, GR203040 produced a rightward displacement of the concentration-effect curves to substance P methyl ester (SPOMe) with suppression of the maximum agonist response (apparent pKB values of 11.9, 11.2 and 11.1 respectively). 5. In anaesthetized rabbits, GR203040

antagonized reductions in carotid arterial vascular resistance evoked by SPOMe, injected via the lingual artery (DR10 (i.e. the dose producing a dose-ratio of 10) = 1.1 micrograms kg-1, i.v.). At a dose 20 fold greater than its DR10 value (i.e. 22 micrograms kg-1, i.v.), significant antagonism was evident more than 2 h after GR203040 administration. 6. In anaesthetized rats, GR203040 (3 and 10 mg kg-1, i.v.) produced a dose-dependent inhibition of plasma protein extravasation in dura mater, conjunctiva, eyelid and lip in response to electrical stimulation of the trigeminal ganglion. 7. It is concluded that GR203040 is one of the most potent and selective NK1 receptor antagonists yet described, and as such, has considerable potential as a pharmacological tool to characterize the physiological and pathological roles of substance P and NK1 receptors. GR203040 may also have potential as a novel therapeutic agent for the treatment of conditions such as migraine, emesis and pain.

L10 ANSWER 57 OF 68 MEDLINE on STN DUPLICATE 22
ACCESSION NUMBER: 95382788 MEDLINE
DOCUMENT NUMBER: PubMed ID: 7654208
TITLE: Purification and characterization of a fatty acid-activated protein kinase (**PKN**) from rat testis.
AUTHOR: Kitagawa M; Mukai H; Shibata H; Ono Y
CORPORATE SOURCE: Department of Biology, Faculty of Science, Kobe University, Japan.
SOURCE: Biochemical journal, (1995 Sep 1) 310 (Pt 2) 657-64. Journal code: 2984726R. ISSN: 0264-6021.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199509
ENTRY DATE: Entered STN: 19951005
Last Updated on STN: 19970203
Entered Medline: 19950927

AB **PKN**, a novel protein kinase with a catalytic domain homologous to that of the protein kinase C (PKC) family and unique N-terminal leucine-zipper-like sequences, was identified by molecular **cloning** from a **human** hippocampus cDNA library [Mukai and Ono (1994) Biochem. Biophys. Res. Commun. 199, 897-904]. Recently we partially purified **recombinant PKN** from COS7 cells transfected with the cDNA construct encoding **human PKN**, and demonstrated that the **recombinant PKN** was activated by unsaturated fatty acids and limited proteolysis [Mukai, Kitagawa, Shibata et al. (1994) Biochem. Biophys. Res. Commun. 204, 348-356]. The present work has focused on the further purification and characterization of **PKN** from native rat tissue. Immunochemical measurement revealed that **PKN** was found in every tissue, and was especially abundant in testis, spleen and brain; subcellular fractionation of rat brain showed that half of the **PKN** was localized in the soluble cytosolic fraction. **PKN** was purified approx. 8000-fold to apparent homogeneity from the cytosolic fraction of rat testis by DEAE-cellulose chromatography, ammonium sulphate fractionation and chromatography on butyl-Sepharose, heparin-Sepharose, Mono Q and protamine-CH-Sepharose. The enzyme migrates as a band of apparent molecular mass 120 kDa. Using serine-containing peptides based on the pseudosubstrate sequence of PKC-delta as phosphate acceptors, the kinase activity was stimulated several-fold by 40 microM unsaturated fatty acids or by detergents such as 0.04% sodium deoxycholate and 0.004% SDS. In the absence of modifiers, protamine sulphate, myelin basic protein and synthetic peptides based on the pseudosubstrate site of PKCs or ribosomal S6 protein were good substrates for phosphorylation by the kinase. In the presence of 40 microM arachidonic acid the kinase activity of **PKN** for these phosphate acceptors was increased 2-18-fold. The autophosphorylation activity of purified **PKN** was partially inhibited by pretreatment with alkaline phosphatase. These properties

appear to distinguish **PKN** from many protein kinases isolated previously.

L10 ANSWER 58 OF 68 MEDLINE on STN
ACCESSION NUMBER: 95154310 MEDLINE
DOCUMENT NUMBER: PubMed ID: 7851406
TITLE: **Cloning** and **expression** patterns of two members of a novel protein-kinase-C-related kinase family.
AUTHOR: Palmer R H; Ridden J; Parker P J
CORPORATE SOURCE: Protein Phosphorylation Laboratory, Imperial Cancer Research Fund, London, England.
SOURCE: European journal of biochemistry / FEBS, (1995 Jan 15) 227 (1-2) 344-51.
Journal code: 0107600. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-S75546; GENBANK-S75548; GENBANK-U33052; GENBANK-U33053
ENTRY MONTH: 199503
ENTRY DATE: Entered STN: 19950322
Last Updated on STN: 19960315
Entered Medline: 19950313

AB The cDNA **clones** for two members of a novel protein kinase family were isolated and sequenced. These protein-kinase-C-related kinases, PRK1 and PRK2, display extensive identity to each other, revealing non-kinase domain similar regions. HR1 and HR2. HR1 contains a motif repeated three times (HR1a-c), while HR2 shows similarity to the amino-terminal sequence of protein-kinase-C epsilon and eta isotypes. Both PRK1 and PRK2, **expressed** in COS 1 cells, are autophosphorylated in immunoprecipitates, indicating intrinsic kinase activity. PRK1 and PRK2, as well as a third member of this family, PRK3, show distinct patterns of **expression** in adult tissues.

L10 ANSWER 59 OF 68 MEDLINE on STN
ACCESSION NUMBER: 95344386 MEDLINE
DOCUMENT NUMBER: PubMed ID: 7619073
TITLE: **Expression**, purification and characterization of the ubiquitous protein kinase C-related kinase 1.
AUTHOR: Palmer R H; Parker P J
CORPORATE SOURCE: Imperial Cancer Research Fund, London, U.K.
SOURCE: Biochemical journal, (1995 Jul 1) 309 (Pt 1) 315-20.
Journal code: 2984726R. ISSN: 0264-6021.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199508
ENTRY DATE: Entered STN: 19950905
Last Updated on STN: 19970203
Entered Medline: 19950818

AB The recently described protein kinase C-related kinase (PRK) family is comprised of at least three members: PRK1, PRK2 and PRK3. Here the **expression**, purification and characterization of the ubiquitously **expressed** isoform, PRK1, is described. The enzyme was **expressed** in COS 7 cells and subsequently purified to apparent homogeneity by sequential column chromatography. The purified PRK1 protein migrates as a single 120 kDa polypeptide on SDS/PAGE. It displays a substrate specificity that in part resembles that of protein kinase C (PKC); however, unlike PKC, it is not activated by any combination of phorbol esters, diacylglycerol and Ca2+. Nevertheless, it can be activated by limited proteolysis, indicating a negative regulatory role for the N-terminal domain(s). PRK1 is also activated by phospholipids.

The physiological relevance of this activation is discussed.

L10 ANSWER 60 OF 68 MEDLINE on STN DUPLICATE 23
ACCESSION NUMBER: 95226461 MEDLINE
DOCUMENT NUMBER: PubMed ID: 7711077
TITLE: Xenopus **PKN**: **cloning** and sequencing of the cDNA and identification of conserved domains.
AUTHOR: Mukai H; Mori K; Takanaga H; Kitagawa M; Shibata H; Shimakawa M; Miyahara M; Ono Y
CORPORATE SOURCE: Department of Biology, Faculty of Science, Kobe University, Japan.
SOURCE: Biochimica et biophysica acta, (1995 Apr 4) 1261 (2) 296-300.
Journal code: 0217513. ISSN: 0006-3002.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-D43890
ENTRY MONTH: 199505
ENTRY DATE: Entered STN: 19950524
Last Updated on STN: 19950524
Entered Medline: 19950515
AB cDNA **clone** encoding Xenopus laevis **PKN** has been isolated from Xenopus kidney library. Sequencing of this **clone** has revealed a single open reading frame encoding a protein of 901 amino acids. Immunoprecipitate from cytoplasmic fraction of COS7 cells transfected with this cDNA construct using antiserum against bacterially **expressed** Xenopus **PKN** revealed arachidonic acid-dependent autophosphorylation activity. Comparison of the closely related sequences of **human** and rat **PKN** with a protein from evolutionarily distant Xenopus, revealed several highly invariant domains in the NH2-terminal regulatory regions, suggesting that they participate in binding interaction with arachidonic acid.

L10 ANSWER 61 OF 68 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
ACCESSION NUMBER: 95:258925 SCISEARCH
THE GENUINE ARTICLE: QQ952
TITLE: XENOPUS **PKN** - **CLONING** AND SEQUENCING OF THE CDNA AND IDENTIFICATION OF CONSERVED DOMAINS
AUTHOR: MUKAI H; MORI K; TAKANAGA H; KITAGAWA M; SHIBATA H; SHIMAKAWA M; MIYAHARA M; ONO Y (Reprint)
CORPORATE SOURCE: KOBE UNIV, FAC SCI, DEPT BIOL, KOBE 657, JAPAN (Reprint); KOBE UNIV, FAC SCI, DEPT BIOL, KOBE 657, JAPAN
COUNTRY OF AUTHOR: JAPAN
SOURCE: BIOCHIMICA ET BIOPHYSICA ACTA-GENE STRUCTURE AND EXPRESSION, (04 APR 1995) Vol. 1261, No. 2, pp. 296-300.
ISSN: 0167-4781.
DOCUMENT TYPE: Note; Journal
FILE SEGMENT: LIFE
LANGUAGE: ENGLISH
REFERENCE COUNT: 6
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB cDNA **clone** encoding Xenopus laevis **PKN** has been isolated from Xenopus kidney library. Sequencing of this **clone** has revealed a single open reading frame encoding a protein of 901 amino acids. Immunoprecipitate from cytoplasmic fraction of COS7 cells transfected with this cDNA construct using antiserum against bacterially **expressed** Xenopus **PKN** revealed arachidonic acid-dependent autophosphorylation activity. Comparison of the closely related sequences of **human** and rat **PKN** with a protein from evolutionarily distant Xenopus, revealed several highly invariant domains in the NH2-terminal regulatory regions, suggesting that they participate in binding interaction with arachidonic acid.

L10 ANSWER 62 OF 68 MEDLINE on STN DUPLICATE 24
ACCESSION NUMBER: 94183274 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8135837
TITLE: A novel protein kinase with leucine zipper-like sequences: its catalytic domain is highly homologous to that of protein kinase C.
AUTHOR: Mukai H; Ono Y
CORPORATE SOURCE: Department of Biology, Faculty of Science, Kobe University, Japan.
SOURCE: Biochemical and biophysical research communications, (1994 Mar 15) 199 (2) 897-904.
Journal code: 0372516. ISSN: 0006-291X.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-D26180; GENBANK-D26181
ENTRY MONTH: 199404
ENTRY DATE: Entered STN: 19940428
Last Updated on STN: 19960129
Entered Medline: 19940421

AB A novel protein kinase, designated **PKN**, was identified by molecular **cloning** from a **human** hippocampus cDNA library. **PKN** consists of 942 amino acids with a calculated molecular mass of 103,925 daltons. **PKN** has leucine zipper-like sequences in its amino terminal region and contains a catalytic domain that shows strong similarity to that of protein kinase C family. Northern blot analysis indicates **PKN** is **expressed** ubiquitously in **human** tissues. Antisera against **PKN** identified a 120K dalton protein on SDS polyacrylamide gel electrophoresis when **PKN** was **expressed** in the insect cells or COS7 cells. **Recombinant PKN** revealed an intrinsic protein kinase activity associated with a 120K protein. This activity was abolished by mutation of the lysine residue in the potential ATP binding site.

L10 ANSWER 63 OF 68 MEDLINE on STN DUPLICATE 25
ACCESSION NUMBER: 95032119 MEDLINE
DOCUMENT NUMBER: PubMed ID: 7945381
TITLE: Activation of **PKN**, a novel 120-kDa protein kinase with leucine zipper-like sequences, by unsaturated fatty acids and by limited proteolysis.
AUTHOR: Mukai H; Kitagawa M; Shibata H; Takanaga H; Mori K; Shimakawa M; Miyahara M; Hirao K; Ono Y
CORPORATE SOURCE: Department of Biology, Faculty of Science, Kobe University, Japan.
SOURCE: Biochemical and biophysical research communications, (1994 Oct 14) 204 (1) 348-56.
Journal code: 0372516. ISSN: 0006-291X.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199411
ENTRY DATE: Entered STN: 19941222
Last Updated on STN: 19970203
Entered Medline: 19941123

AB **PKN**, a novel protein kinase with catalytic domain homologous to PKC family and unique amino terminal leucine zipper-like sequences, was purified partially from COS7 cells transfected with the cDNA construct encoding **human PKN** for enzymatic characterization of the enzyme. Using serine containing synthetic peptides based on PKC pseudosubstrate sites as the phosphate acceptors, kinase activities estimated from partially purified **PKN** were not stimulated by

Ca2+/phosphatidylserine/diolein but were activated several-fold to several tens-fold by 40 microM unsaturated fatty acids, such as arachidonic acid, linoleic acid, and oleic acid. Autophosphorylation of the immunoprecipitates using anti-**PKN** antiserum was also stimulated by various unsaturated fatty acids. Limited proteolysis of **PKN** with trypsin induced an enhancement of the peptide kinase activity that was almost independent of arachidonic acid.

L10 ANSWER 64 OF 68 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1993:412749 BIOSIS
DOCUMENT NUMBER: PREV199396078474
TITLE: Calcium regulation of vasoactive intestinal polypeptide mRNA abundance in SH-SY5Y **human** neuroblastoma cells.
AUTHOR(S): Adler, E. M.; Fink, J. Stephen [Reprint author]
CORPORATE SOURCE: Molecular NMeurobiol. Lab., Massachusetts General Hosp., Boston, MA 02114, USA
SOURCE: Journal of Neurochemistry, (1993) Vol. 61, No. 2, pp. 727-737.
CODEN: JONRA9. ISSN: 0022-3042.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 8 Sep 1993
Last Updated on STN: 3 Jan 1995
AB Second messenger regulation of gene **expression** provides a mechanism by which neurons can transduce environmental stimuli into long-term changes in the **expression** of molecules involved in neuronal signaling. We have investigated calcium-dependent induction of vasoactive intestinal polypeptide (VIP) mRNA and compared it with induction of VIP mRNA by cyclic AMP. Depolarization with 60 mM KCl or exposure to the calcium ionophore A23187 increases VIP mRNA levels in SH-SY5Y cells. The increase in VIP mRNA content in response to Ca-2+ mobilization is slow, independent of adenylate cyclase activation, and requires de novo protein synthesis. The increase in VIP mRNA content in response to elevation of cyclic AMP levels by forskolin/isobutylmethylxanthine is independent of Ca-2+ influx and does not require new protein synthesis. The mRNA for the transcription factors ATF-3, c-fos, c-jun, junB, and zif/268 is induced by A23187. Of these, ATF-3 showed the greatest relative induction by A23187 compared with induction by forskolin/isobutylmethylxanthine.

L10 ANSWER 65 OF 68 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1992:250994 HCAPLUS
DOCUMENT NUMBER: 116:250994
TITLE: Identification of the double-stranded RNA-binding domain of the **human** interferon-inducible protein kinase
AUTHOR(S): Patel, Rekha C.; Sen, Ganes C.
CORPORATE SOURCE: Dep. Mol. Biol., Cleveland Clin. Found., Cleveland, OH, 44195, USA
SOURCE: Journal of Biological Chemistry (1992), 267(11), 7671-6
CODEN: JBCHA3; ISSN: 0021-9258
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The interferon-inducible double-stranded (ds) RNA-activated protein kinase (protein p68 kinase; protein kinase DAI) (I) is a physiol. important enzyme that regulates the rate of cellular and viral protein synthesis by phosphorylating and thereby inactivating protein formation initiation factor 2. A cDNA **clone** of **human** I was investigated by polymerase chain reaction **cloning** using the recently published sequence of this enzyme. Activat I was synthesized by in vitro transcription-translation of the cDNA **clone**. This system was used for mapping the dsRNA-binding domain of I. Progressive deletions

from the C-terminus were introduced by digesting the cDNA with suitable restriction endonucleases. The **expression** of proteins harboring deletions from the N-terminus was achieved by **cloning** DNA fragments into appropriately constructed **expression** vectors. The affinity of the truncated proteins for dsRNA was examined by testing their capacity to bind to dsRNA-agarose beads. The results demonstrated that the dsRNA-binding domain lies at the N-terminus of the protein. A truncated protein containing the 1st 170 amino acid residues from the I N-terminus could bind to dsRNA. However, deletion of 34 residues from the N-terminus or 41 residues from the C-terminus of this truncated protein eliminated its dsRNA-binding activity. Comparison of the primary structure and the secondary structure of this region of I and the corresponding region of 2',5'-oligoadenylate synthetase revealed no apparent similarity.

L10 ANSWER 66 OF 68 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1992:526632 BIOSIS
DOCUMENT NUMBER: PREV199294134707; BA94:134707
TITLE: TAU PROTEIN INDUCES BUNDLING OF MICROTUBULES IN-VITRO COMPARISON OF DIFFERENT TAU ISOFORMS AND A TAU PROTEIN FRAGMENT.
AUTHOR(S): SCOTT C W [Reprint author]; KLIKA A B; LO M M S; NORRIS T E; CAPUTO C B
CORPORATE SOURCE: ICI PHARMACEUTICALS GROUP, ICI AMERICAS INC, LW-215, CONCORD PIKE, WILMINGTON, DEL 19897, USA
SOURCE: Journal of Neuroscience Research, (1992) Vol. 33, No. 1, pp. 19-29.
CODEN: JNREDK. ISSN: 0360-4012.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 19 Nov 1992
Last Updated on STN: 20 Nov 1992

AB **Expression** of tau protein in non-neuronal cells can result in a redistribution of the microtubule cytoskeleton into thick bundles of tau-containing microtubules (Lewis et al: Nature 342:498-505, 1989; Kanai et al: J Cell Biol 109:1173-1184, 1989). We reconstituted microtubule bundles using purified tubulin and tau in order to study the assembly of these structures. Taxol-stabilized tubulin polymers were incubated with various concentrations of **recombinant human** tau and examined by electron microscopy. With increasing concentrations of τ3 (tau isoform containing three microtubule binding domains) or τ4 (isoform containing four microtubule binding domains) the microtubules changed orientation from a random distribution to loosely and tightly packed parallel arrays and then to thick cables. In contrast, τ4L, the tau isoform containing four microtubule binding domains plus a 58-amino acid insert near the N-terminus, showed minimal bundling activity. τ4-induced bundling could be inhibited by the addition of 0.5 M NaCl or 0.4 mM estramustine phosphate, conditions which are known to inhibit tau binding to microtubules. A tau construct that contained only the microtubule binding domains plus 19 amino acids to the C-terminus was fully capable of bundling microtubules. Phosphorylation of τ3 with cAMP-dependent protein kinase had no effect on its ability to induce microtubule bundling. These results indicate that tau protein is directly capable of bundling microtubules in vitro, and suggests that different tau isoforms differ in their ability to bundle microtubule filaments.

L10 ANSWER 67 OF 68 MEDLINE on STN
ACCESSION NUMBER: 90216686 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2324095
TITLE: Positive and negative regulation of a tumor necrosis factor response in melanoma cells.
AUTHOR: Johnson S E; Baglioni C
CORPORATE SOURCE: Department of Biological Sciences, State University of New

York, Albany 12222.
CONTRACT NUMBER: CA-29895 (NCI)
SOURCE: Journal of biological chemistry, (1990 Apr 25) 265 (12) 6642-9.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199005
ENTRY DATE: Entered STN: 19900622
Last Updated on STN: 19970203
Entered Medline: 19900524

AB Tumor necrosis factor (TNF) elicits a wide variety of responses in target cells by binding to cell surface receptors, but the signal transduced from these receptors in unclear. We examined the role of two different second messenger systems in the regulation of plasminogen activator inhibitor, type 2 (PAI-2) induction by TNF in SK-MEL-109 melanoma cells. Synthesis of PAI-2 and transcription of its mRNA could be induced by a protein kinase C (PKC) activator, phorbol myristate acetate. In addition, induction of PAI-2 synthesis by TNF was blocked by two PKC inhibitors, staurosporine and 1-(5-isoquinolinesulfonyl)-2-methylpiperazine dihydrochloride. The inhibitor of cyclic nucleotide-dependent **protein kinases**, **N**-[2-(methylamino)-ethyl]-5-isoquinoline sulfonamide dihydrochloride, was much less effective in decreasing PAI-2 synthesis. Staurosporine and 1-(5-isoquinolinesulfonyl)-2-methylpiperazine dihydrochloride also inhibited both TNF- and phorbol myristate acetate-induced PAI-2 mRNA accumulation. We measured the binding of 3H-labeled phorbol dibutyrate to membrane and cytosol fractions of TNF-treated SK-MEL-109 cells and found a transient redistribution of 3H-labeled phorbol dibutyrate binding from cytosol to membrane fractions in response to TNF. In contrast to the positive regulation by PKC in promoting TNF-induced PAI-2 synthesis cAMP inhibited this response. Pretreatment of cells with agents that raise intracellular cAMP levels completely abolished TNF-induced PAI-2 synthesis. Addition of cAMP-elevating agents during TNF induction could also block PAI-2 synthesis. PAI-2 mRNA accumulation in response to TNF was inhibited, but not completely abolished, by cAMP-elevating agents, suggesting that cAMP also exerted its inhibitory effect at the translation level. The positive regulation of a TNF response by PKC and its negative modulation by cAMP may provide a means for intracellular coordination of signals from interacting extracellular factors in regulating TNF responses in different target cells.

L10 ANSWER 68 OF 68 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
DUPLICATE 26
ACCESSION NUMBER: 1990-02508 BIOTECHDS
TITLE: Effects of temperature on Escherichia coli overproducing beta-lactamase or **human** epidermal growth factor; **recombinant** protein production and secretion
AUTHOR: Chalmers J J; Kim E; Telford J N; Wong E Y; Tacon W T; *Wilson D B
CORPORATE SOURCE: Monsanto
LOCATION: Section of Biochemistry, Molecular and Cell Biology, Cornell University, Ithaca, New York 14853, USA.
SOURCE: Appl.Environ.Microbiol.; (1990) 56, 1, 104-11
CODEN: AEMIDF
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The effects of temperature on Escherichia coli strains which overproduce and excrete beta-lactamase (b-Lase, EC-3.5.2.6) or **human** epidermal growth factor (hEGF) were studied. E. coli RB791(lacIq) was used as a host. Plasmid **pKN** was pBR322 modified by placement of the tac promoter upstream of the gene for b-Lase. The tac promoter was induced

by isopropyl-beta-thiogalactoside (IPTG). E. coli cells containing plasmid **pKN** were grown in batch culture at 37, 30, 25 and 20 deg. The amount of active b-Lase increased with decreased temperature; purity of the enzyme increased at lower temperature (45% at 37 deg to 90% at 20 deg). Continuous culture at 37 and 30 deg was difficult due to poor cell reproduction and b-Lase production. However, at 20 deg, continuous production and excretion of b-Lase was obtained for more than 450 hr. When E. coli RB791 cells carrying plasmid pUC encoding hEGF were grown at 31 and 37 deg, significant cell lysis occurred. However, almost all cells were intact at 21 and 25 deg. A specific productivity of 70 ug hEGF/mg total protein was obtained at 21 deg. Decreased growth temperature may be important for the production of some plasmid-encoded proteins. (24 ref)

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E2            2     RUSCH CRAIG D/AU
E3          161 --> RUSCH D/AU
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E5           17     RUSCH D B/AU
E6          113     RUSCH D H/AU
E7            3     RUSCH D N/AU
E8            7     RUSCH D T/AU
E9          175     RUSCH D W/AU
E10           3     RUSCH DANA/AU
E11           1     RUSCH DAVID/AU
E12           2     RUSCH DAVID N/AU

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L11         161 "RUSCH D"/AU

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E3          221 --> KETCHUM K A/AU
E4            1     KETCHUM K J/AU
E5           32     KETCHUM K L/AU
E6           20     KETCHUM KAREN/AU
E7          179     KETCHUM KAREN A/AU
E8            1     KETCHUM KAREN ANN/AU
E9            2     KETCHUM KATHY/AU
E10           4     KETCHUM KEVIN/AU
E11           3     KETCHUM KEVIN L/AU
E12           2     KETCHUM KRISTY/AU

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                "KETCHUM KAREN"/AU OR "KETCHUM KAREN A"/AU)

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E2            1     DIFRANCESCO U M/AU
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E5            1     DIFRANCESCOL/AU
E6            1     DIFRANCESO D/AU
E7            2     DIFRANCESO L/AU
E8            1     DIFRANCESO R/AU
E9            1     DIFRANCESO ROBIN/AU
E10           1     DIFRANCESSO L/AU
E11           6     DIFRANCIA C/AU
E12           4     DIFRANCIA CELENE/AU
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L13         112 ("DIFRANCESCO V"/AU OR "DIFRANCESCO VALENTINA"/AU)

=> s beasley e m/au
L14         298 BEASLEY E M/AU

=> d his

     (FILE 'HOME' ENTERED AT 14:42:22 ON 16 JUL 2004)

     FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS,
     LIFESCI' ENTERED AT 14:43:13 ON 16 JUL 2004
L1          1044 S "PROTEIN KINASE N" OR "PKN"
L2           226 S HUMAN AND L1
L3       6609293 S CLON? OR EXPRESS? OR RECOMBINANT
L4           141 S L2 AND L3
L5          1645 S RETINOBLASTOMAS OR PLACENTA(A)CHORIOCARCINOMAS OR BOCIO (A)TU
L6         66422 S LEUKEMIAS OR (WILM (2W) TUMOR?) OR "BRAIN (A) ANAPLASTIC(A)OL
L7         67972 S L5 OR L6
L8             2 S L4 AND L7
L9             2 DUP REM L8 (0 DUPLICATES REMOVED)
L10           68 DUP REM L4 (73 DUPLICATES REMOVED)
                 E RUSCH D/AU
L11           161 S E3
                 E KETCHUM K A/AU
L12           453 S E3-E7
                 E DIFRANCESCO V/AU
L13           112 S E3-E4
L14           298 S BEASLEY E M/AU

=> s l11 or l12 or l13 or l14
L15         897 L11 OR L12 OR L13 OR L14

=> s l2 and l15
L16           5 L2 AND L15

=> dup rem l16
PROCESSING COMPLETED FOR L16
L17           5 DUP REM L16 (0 DUPLICATES REMOVED)

=> d 1-5 ibib ab

L17  ANSWER 1 OF 5  BIOSIS  COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER:   2004:28895  BIOSIS
DOCUMENT NUMBER:    PREV200400030062
TITLE:              Isolated human kinase proteins.
AUTHOR(S):          Wei, Ming-Hui [Inventor, Reprint Author];
                    Chandramouliswara, Ishwar [Inventor]; Ye, Jane [Inventor];
                    Ketchum, Karen A. [Inventor]; Di Francesco,
                    Valentina [Inventor]; Beasley, Ellen M. [Inventor]
CORPORATE SOURCE:   Silver Spring, MD, USA
                    ASSIGNEE: Applera Corporation
PATENT INFORMATION: US 6649389 November 18, 2003
SOURCE:             Official Gazette of the United States Patent and Trademark
                    Office Patents, (Nov 18 2003) Vol. 1276, No. 3.
                    http://www.uspto.gov/web/menu/patdata.html. e-file.
                    ISSN: 0098-1133 (ISSN print).
DOCUMENT TYPE:      Patent
LANGUAGE:           English
ENTRY DATE:         Entered STN: 31 Dec 2003
                    Last Updated on STN: 31 Dec 2003
AB   The present invention provides amino acid sequences of peptides that are
     encoded by genes within the human genome, the kinase peptides of
     the present invention.  The present invention specifically provides
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isolated peptide and nucleic acid molecules, methods of identifying orthologs and paralogs of the kinase peptides, and methods of identifying modulators of the kinase peptides.

L17 ANSWER 2 OF 5 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:594987 HCAPLUS
DOCUMENT NUMBER: 137:151129
TITLE: Protein, gene and cDNA sequences of a novel **human** protein kinase related to protein kinase **PKN** subfamily and their uses in drug screening
INVENTOR(S): Rusch, Douglas; **Ketchum, Karen A.**; Di Francesco, Valentina; Beasley, Ellen M.
PATENT ASSIGNEE(S): PE Corporation, USA
SOURCE: PCT Int. Appl., 76 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002061062	A2	20020808	WO 2002-US2152	20020129
WO 2002061062	A3	20030522		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
US 6500655	B1	20021231	US 2001-849334	20010507
EP 1358338	A2	20031105	EP 2002-713461	20020129
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				

PRIORITY APPLN. INFO.:
US 2001-773371 A 20010201
US 2001-849334 A 20010507
WO 2002-US2152 W 20020129

AB The invention provides protein, cDNA and genomic sequences for a novel **human** protein kinase related to protein kinase **PKN** subfamily. The protein kinase gene is expressed in **human** eye retinoblastomas, placenta choriocarcinomas, germ cells, bocio tumors, pre-B cell acute lymphoblastic leukemias, wilm's tumors of the kidney, uterus tumors, brain anaplastic oligodendromas, uterus endometrial adenocarcinomas, and leukocytes. The protein kinase gene has been mapped to chromosome 8. The invention also relates to screening modulator of said protein kinase and use them in therapy. The invention further relates to methods, vector and hosts for expression of said protein kinase.

L17 ANSWER 3 OF 5 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:409190 HCAPLUS
DOCUMENT NUMBER: 137:1566
TITLE: Protein, gene and cDNA sequences of a novel **human protein kinase N** sequence homolog
INVENTOR(S): Wei, Ming-hui; Chandramouliswaran, Ishwar; Ye, Jane; **Ketchum, Karen A.**; Di Francesco, Valentina; Beasley, Ellen M.
PATENT ASSIGNEE(S): USA
SOURCE: U.S. Pat. Appl. Publ., 39 pp., Cont.-in-part of U.S.

Ser. No. 734,032.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2002064851	A1	20020530	US 2001-816094	20010326
US 6534299	B2	20030318		
US 2002103116	A1	20020801	US 2000-734032	20001212
WO 2001088148	A2	20011122	WO 2001-US15776	20010517
WO 2001088148	A3	20031016		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
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EP 1373516	A2	20040102	EP 2001-952118	20010517
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
JP 2004507223	T2	20040311	JP 2001-584530	20010517
US 2003022339	A1	20030130	US 2002-233613	20020904
US 6649389	B2	20031118		
US 2004038362	A1	20040226	US 2003-639429	20030813

PRIORITY APPLN. INFO.:

US 2000-205228P	P	20000517
US 2000-734032	A2	20001212
US 2001-816094	A	20010326
WO 2001-US15776	W	20010517
US 2002-233613	A3	20020904

AB The invention provides protein, cDNA and genomic sequences for a novel **human** protein, which shares sequence homol. to a known kinase and is related to the **protein kinase N** subfamily. The kinase sequence homolog gene is expressed in **humans** in the brain, placenta, kidney and heart. Seven one novel single nucleotide polymorphism sites (beyond the ORF or in intron regions), including three indels, have been identified on kinase sequence homolog gene. Thus, the present invention specifically provides isolated protein and nucleic acid mols., methods of identifying orthologs and paralogs of the kinases, methods of identifying modulators of the kinases, and methods of diagnosis and treatment of diseases associated with the kinase.

L17 ANSWER 4 OF 5 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2003:1236 HCAPLUS
DOCUMENT NUMBER: 138:68934
TITLE: Identification, genomic and cDNA sequences and cloning of a **human protein kinase N** sequence homolog
INVENTOR(S): Rusch, Douglas; **Ketchum, Karen A.**; Di Francesco, Valentina; Beasley, Ellen M.
PATENT ASSIGNEE(S): Applera Corporation, USA
SOURCE: U.S., 44 pp., Cont.-in-part of U. S. Ser. No. 773,371, abandoned.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6500655	B1	20021231	US 2001-849334	20010507
WO 2002061062	A2	20020808	WO 2002-US2152	20020129
WO 2002061062	A3	20030522		

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

EP 1358338	A2	20031105	EP 2002-713461	20020129

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

WO 2002090525	A2	20021114	WO 2002-US7155	20020308
WO 2002090525	A3	20030327		

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

EP 1385863	A2	20040204	EP 2002-725095	20020308

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

US 2003049792	A1	20030313	US 2002-274878	20021022
US 6670163	B2	20031230		
US 2004067522	A1	20040408	US 2003-697266	20031031

PRIORITY APPLN. INFO.:

US 2001-773371	B2	20010201
US 2001-849334	A	20010507
WO 2002-US2152	W	20020129
WO 2002-US7155	W	20020308
US 2002-274878	A3	20021022

AB The present invention is based in part on the identification of amino acid sequences of **human** kinase peptides and proteins that are related to the **protein kinase N** (**PKN**) subfamily, as well as allelic variants and other mammalian orthologs thereof. The present invention provides genomic, cDNA and amino acid sequences of the **human protein kinase N** sequence homolog. Chromosomal mapping of the **protein kinase N** sequence homolog gene, tissue-specific expression profiles, and structural motifs of the polypeptides are provided. The protein and nucleic acid sequences of the invention, can be used as models for the development of **human** therapeutic targets, aid in the identification of therapeutic proteins, and serve as targets for the development of **human** therapeutic agents that modulate kinase activity in cells and tissues that express the kinase. Expression of the **protein kinase N** sequence homolog gene in **humans** in eye retinoblastomas, placenta choriocarcinomas, germ cells, bocio tumors, pre-B cell acute lymphoblastic leukemias, Wilm's tumors of the kidney, uterus tumors, brain anaplastic oligodendromas, uterus endometrial adenocarcinomas, and leukocytes is reported.

REFERENCE COUNT: 1 THERE ARE 1 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L17 ANSWER 5 OF 5 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
ACCESSION NUMBER: 2002-07405 BIOTECHDS
TITLE: **Human** kinase protein and polynucleotides encoding them, useful for identifying modulators of kinase polypeptides and for treating, preventing, and/or diagnosing neurodegenerative diseases and cancer; vector-mediated recombinant protein gene transfer and expression in host cell, DNA probe, antibody, DNA chip and transgenic animal for disease prevention, diagnosis and gene therapy
AUTHOR: WEI M; CHANDRAMOULISWARA I; YE J; **KETCHUM K A**; DI FRANCESCO V; **BEASLEY E M**
PATENT ASSIGNEE: APPLERA CORP
PATENT INFO: WO 2001088148 22 Nov 2001
APPLICATION INFO: WO 2000-US15776 17 May 2000
PRIORITY INFO: US 2001-816094 26 Mar 2001
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-089857 [12]
AB DERWENT ABSTRACT:

NOVELTY - An isolated protein (a member of kinase family of protein and is related to **PKN** kinase subfamily) consisting or comprising a fully defined sequence of 424 amino acids (S2) as given in the specification, or its fragment comprising 10 contiguous amino acids, or an amino acid sequence of an allelic variant or ortholog of the amino acid sequence of (S2), is new.

DETAILED DESCRIPTION - An isolated protein (a member of kinase family of protein and is related to **PKN** kinase subfamily) consisting or comprising a fully defined sequence of 424 amino acids (S2) as given in the specification, or its fragment comprising 10 contiguous amino acids, or an amino acid sequence of an allelic variant or ortholog of the amino acid sequence of (S2), is new. (I) consists of or comprises: an amino acid sequence of (S2); an amino acid sequence of an allelic variant or an ortholog of (S2), where the allelic variant or ortholog is encoded by a nucleic acid molecule that hybridizes under stringent conditions to the opposite strand of a nucleic acid molecule having a fully defined sequence of 2598 nucleotides (S1) (transcript/cDNA) or 7301 nucleotides (S3) (genomic DNA) as given in the specification; a fragment of an amino acid sequence of (S2), comprising 10 contiguous amino acids. INDEPENDENT CLAIMS are also included for the following: (1) an isolated antibody (II) that selectively binds to (I) comprising the amino acid sequence of (S2), its allelic variant or ortholog, or fragment; (2) an isolated nucleic acid molecule (III) consisting or comprising of a nucleotide sequence that encodes (I) or a nucleotide sequence that is complement of the nucleotide sequence encoding (I); (3) a gene chip comprising (III) that comprises a nucleotide sequence encoding (I), or its complement; (4) a transgenic non-**human** animal comprising (III) that comprises a nucleotide sequence encoding (I), or its complement; (5) a nucleic acid vector (IV) comprising (III) that comprises a nucleotide sequence encoding (I), or its complement; (6) a host cell comprising (IV); (7) preparation of (I); (8) detecting the presence of (I) comprising the amino acid sequence of (S2), its allelic variant or ortholog, or fragment, in a sample involves contacting the sample with a detection agent that specifically allows detection of the presence of the peptide in the sample and then detecting the presence of the peptide; (9) detecting the presence of (III) in a sample involves contacting the sample with an oligonucleotide that hybridizes to the nucleic acid molecule under stringent conditions and determining whether a oligonucleotide binds to the nucleic acid molecule in the sample; (10) a pharmaceutical composition (V) comprising an agent that binds to (I) comprising an amino acid sequence of (S2), its allelic variant or ortholog or fragment, and a carrier; (10) an isolated **human** kinase peptide (VI) having an amino acid sequence that shares 70% homology with (S2); and (11) an isolated nucleic acid molecule (VII)

encoding a **human** kinase peptide which shares at least 80% homology with (S1) or (S3).

WIDER DISCLOSURE - The following are disclosed: (1) isolated peptide and protein molecules that consist essentially of the amino acid sequence of (S2), its allelic variant or ortholog, or fragment; (2) nucleic acid molecules that consist essentially of nucleotide sequence that encodes (I) or a nucleotide sequence that is complement of the nucleotide sequence encoding (I); (3) chimeric or fusion proteins comprising (I); (4) derivatives or analogs of (I) in which a substituted amino acid residue is not one encoded by the genetic code; (5) paralogs of the kinase polypeptide; (6) novel agents identified by the above mentioned screening methods;(7) kit comprising (II) for detecting (I) comprising an amino acid sequence of (S2), its allelic variant or ortholog or fragment; (8) non-coding fragments of a nucleic acid molecule having a sequence of (S1) or (S3); and (9) kits for detecting the presence of kinase protein nucleic acid in a biological sample.

BIOTECHNOLOGY - Preparation: (I) is prepared by standard recombinant techniques (claimed). Preferred Molecules: (VI) shares 90% homology with (S2), and (VII) shares at least 90% homology with (S1) or (S3).

ACTIVITY - Cytostatic; neuroprotective.

MECHANISM OF ACTION - Gene therapy; **human** kinase protein expression or activity modulator. No supporting data is given.

USE - The nucleic acids and polypeptides may be used in the prevention, diagnosis and treatment of diseases associated with inappropriate kinase expression. For example, the nucleic acids (or vectors containing them) and the kinase may be used to treat disorders associated with decreased expression by rectifying mutations or deletions in a patient's genome that affect the activity of the enzyme by expressing inactive proteins or to supplement the patients own production of kinases. Additionally, the nucleic acids may be used to produce the kinase, by inserting the nucleic acids into a host cell and culturing the cell to express the protein. The nucleic acid and its complementary sequences may also be used as DNA probes in diagnostic assays to detect and quantitate the presence similar nucleic acids in samples, and therefore which patients may be in need of restorative therapy. The polypeptides may also be used as antigens in the production of antibodies against the kinase and in assays to identify modulators of kinase expression and activity. The anti-kinase antibodies and antagonists may also be used to down regulate expression and activity. The anti-kinase antibodies may also be used as diagnostic agents for detecting the presence of kinase polypeptides in samples (e.g. by enzyme linked immunosorbant assay (ELISA)). Disorders that may be prevented, diagnosed and/or treated by the above methods include, for example neurodegenerative diseases.

ADMINISTRATION - No specific administration details are given.

EXAMPLE - None given.(65 pages)

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(FILE 'HOME' ENTERED AT 14:42:22 ON 16 JUL 2004)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 14:43:13 ON 16 JUL 2004

L1	1044	S	"PROTEIN KINASE N" OR "PKN"
L2	226	S	HUMAN AND L1
L3	6609293	S	CLON? OR EXPRESS? OR RECOMBINANT
L4	141	S	L2 AND L3
L5	1645	S	RETINOBLASTOMAS OR PLACENTA(A)CHORIOCARCINOMAS OR BOCIO (A)TU
L6	66422	S	LEUKEMIAS OR (WILM (2W) TUMOR?) OR "BRAIN (A) ANAPLASTIC(A)OL
L7	67972	S	L5 OR L6
L8	2	S	L4 AND L7
L9	2		DUP REM L8 (0 DUPLICATES REMOVED)
L10	68		DUP REM L4 (73 DUPLICATES REMOVED)

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                E RUSCH D/AU
L11         161 S E3
                E KETCHUM K A/AU
L12         453 S E3-E7
                E DIFRANCESCO V/AU
L13         112 S E3-E4
L14         298 S BEASLEY E M/AU
L15         897 S L11 OR L12 OR L13 OR L14
L16           5 S L2 AND L15
L17           5 DUP REM L16 (0 DUPLICATES REMOVED)
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	Issue Date	Pages	Document ID	Title
1	20040408	47	US 20040067522 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins and uses thereof
2	20040325	82	US 20040058325 A1	Gene expression in biological conditions
3	20040226	40	US 20040038362 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof
4	20040115	60	US 20040009477 A1	Methods for producing libraries of expressible gene sequences
5	20040108	96	US 20040006212 A1	Antibody and antibody fragments for inhibiting the growth of tumors
6	20040108	53	US 20040005648 A1	PYK2 related products and methods
7	20031204	95	US 20030224001 A1	Antibody and antibody fragments for inhibiting the growth of tumors
8	20030717	102	US 20030134302 A1	Libraries of expressible gene sequences
9	20030626	37	US 20030119067 A1	PYK2 related products and methods

	Issue Date	Pages	Document ID	Title
10	20030417	102	US 20030073163 A1	Libraries of expressible gene sequences
11	20030313	47	US 20030049792 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins and uses thereof
12	20030227	122	US 20030040089 A1	Protein-protein interactions in adipocyte cells
13	20030130	40	US 20030022339 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof
14	20020801	34	US 20020103116 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof
15	20020530	39	US 20020064851 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof
16	20020530	34	US 20020064528 A1	Antibodies specific to KDR and uses thereof
17	20020425	37	US 20020048782 A1	PYK2 RELATED PRODUCTS AND METHODS
18	20031230	44	US 6670163 B2	Isolated human kinase proteins

	Issue Date	Pages	Document ID	Title
19	20031209	79	US 6660837 B1	Modified protein derived from protein kinase N
20	20031118	38	US 6649389 B2	Isolated human kinase proteins
21	20030318	37	US 6534299 B2	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof
22	20021231	44	US 6500655 B1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof
23	20020924		US 6455677 B1	FAP.alpha.-specific antibody with improved producibility
24	19981117	49	US 5837815 A	PYK2 related polypeptide products

	Issue Date	Pages	Document ID	Title
1	20040408	47	US 20040067522 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins and uses thereof
2	20030313	47	US 20030049792 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins and uses thereof
3	20031230	44	US 6670163 B2	Isolated human kinase proteins
4	20021231	44	US 6500655 B1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof

	Issue Date	Pages	Document ID	Title
1	20040408	47	US 20040067522 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins and uses thereof
2	20040226	40	US 20040038362 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof
3	20030313	47	US 20030049792 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins and uses thereof
4	20030130	40	US 20030022339 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof
5	20020801	34	US 20020103116 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof
6	20020530	39	US 20020064851 A1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof
7	20031230	44	US 6670163 B2	Isolated human kinase proteins
8	20031118	38	US 6649389 B2	Isolated human kinase proteins
9	20030318	37	US 6534299 B2	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof

	Issue Date	Pages	Document ID	Title
10	20021231	44	US 6500655 B1	Isolated human kinase proteins, nucleic acid molecules encoding human kinase proteins, and uses thereof

	L #	Hits	Search Text
1	L1	38	"protein kinase N"
2	L2	193	"PKN"
3	L3	206	l1 or l2
4	L4	422500	human
5	L5	38	l3 same l4
6	L6	642986	clon$3 or express$3 or recombinant
7	L7	24	l5 same l6
8	L8	390	retinoblastomas or (palcenta adj choriocarcinomas)
9	L9	8374	bocio adj tumor$2 or "leukemias" or "Wilm's tumor$2"
10	L10	8532	l8 or l9
11	L11	4	l3 same l10
12	L12	5824	BEASLEY DIFRANCESCO RUSCH KETCHUM
13	L13	10	l3 and l12